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Characterization of and Herbivore Impact on the Flora and Funga at the Antisana Hydrological Conservation Area.

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Dedication

I dedicate the efforts documented in this thesis to my family, with special gratitude to my grandparents, Imelda and Guillermo, whose unwavering support and assistance were indispensable throughout the course of my endeavors.

To my beloved mother, Betty Méndez, whose profound appreciation for the wonders of nature has profoundly influenced my own perspective. I attribute all of my achievements to my mother; may they be celebrated in your eternal abode.

To my father, Camilo, whose guidance and example have taught me the value of perseverance, I will always remember “Así se templó el acero.”

To my brother Zaid, whose presence have enriched my journey in countless ways.

To Kat, for her companionship and presence “right here and right now”, which has brought solace and strength during this pursuit.

Bryan Israel Vásquez Méndez

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I express my heartfelt thanks to the faculty members of the School of Biological Sciences and Engineering of Yachay Tech University for imparting upon me the principles of scientific inquiry and equipping me with the necessary tools to undertake a project of this magnitude.

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I am indebted to FONAG and its dedicated staff in ACHA for their financial support, logistical assistance, and instrumental role in establishing the necessary installations for this project. Special appreciation goes to Braulio Lahuatte and Guardapáramo Manuel Simba for their tireless dedication and commitment to the preservation of Antisana Paramo, which exemplifies a profound love for this unique ecosystem translated into diligent labor.

I acknowledge that this project would not have been feasible without the collective support and guidance of all mentioned individuals and institutions.

In the Páramo, we find ourselves immersed in Humboldt's vision, and as we progress, we strive towards a deeper understanding of the truth inherent in nature "Naturwahrheit".

All photographs included in this thesis were taken in ACHA by the author, unless otherwise indicated.

Bryan Israel Vásquez Méndez

Resumen

Esta tesis examina los patrones de dominancia y diversidad de las comunidades de plantas vasculares en el Área de Conservación Hídrica Antisana, fuente vital de agua para Quito. Se establecieron 54 parcelas, de 3 m x 2 m, para evaluar la dominancia de la vegetación se usó el método del cuadrado puntual, se representado los resultados en curvas de rango-abundancia. Este estudio cubre seis categorías de hábitat: vegetación de pajonal, arbustiva, herbácea seca, herbácea húmeda, plantas almohadilla y suelo expuesto. En total, se identificaron 69 especies de traqueófitos, que abarcan 47 géneros y 23 familias. Doce especies fueron endémicas del páramo ecuatoriano, 56 nativas de páramos andinos, y una introducida. La riqueza de especies fue mayor en la categoría de hábitat de vegetación arbustiva (SHR = 23 spp.), y menor en suelo expuesto (EMD = 6 spp.). La diversidad, medida por el Número Efectivo de Especies basado en el Índice de Diversidad de Shannon, varió entre las categorías de hábitat. La parcela más diversa estuvo en vegetación arbustiva (SHR = 13 spp. efectivas), en contraste, la vegetación de pajonal tuvo la diversidad más baja (GMC = 3 spp. efectivas). Además, se identificaron seis especies de cuerpos fructíferos de macrohongos, lo que sugiere la presencia de redes de micelio en todas las categorías de hábitat. Los hongos observados incluyeron patógenos de plantas, saprofitos y micorrizas, los cuales desempeñan roles en el ciclo de nutrientes y en la dinámica de las poblaciones vegetales, con un posible impacto en la regulación hídrica. Algunas especies de hongos fueron muy apetecibles para el venado de cola blanca sugiriendo un vínculo entre la diversidad fúngica y las preferencias de herbívoros. Esta caracterización representa la fase inicial de una investigación de herbivoría con una duración de cuatro años.

Palabras clave:

Dominancia, Diversidad, Plantas vasculares, Número Efectivo de Especies, Cuerpos fructíferos macrofúngicos, Venado de cola blanca, Área de Conservación Hídrica Antisana.

Abstract

This thesis investigates the dominance and diversity of vascular plant communities in the Antisana Hydric Conservation Area, a critical water source for Quito. Fifty-four plots, each 3m x 2m, were established to assess tracheophyte dominance using the Point-Intercept method, with tracheophyte dominance patterns represented in Rank-abundance curves. This study spans six habitats: Grassland vegetation, Shrubby vegetation, Dry Herbaceous vegetation, Humid Herbaceous vegetation, Cushion plants, and Exposed Soil. Across these a total of 69 tracheophyte species were identified, covering 47 genera and 23 families. Twelve species were endemic to the Ecuadorian páramo, the other 56 were native to Andean páramos, and one was classified as introduced. Species richness was highest in the Shrubby vegetation habitat category (SHR = 23 spp.), and the lowest in Exposed soil (EMD = 6 spp.). Diversity, measured by the Effective Number of Species based on the Shannon Diversity Index, varied among habitat categories. The most diverse plot was in Shrubby vegetation (SHR = 13 effective spp.), in contrast, Grassland vegetation had the lowest diversity (GMC = 3 effective spp.). Additionally, six macro-fungal fruiting bodies species were identified, suggesting the presence of mycelial networks in all habitat categories. The observed fungi included plant pathogens, saprophytes and mycorrhizae, which play roles in nutrient cycling and plant population dynamics, potentially impacting water regulation. Some mushroom species were very palatable to white-tailed deer, suggesting a potential link between fungal diversity and herbivore preferences, warranting further analysis for restoration practices. To assess herbivore impacts, exclusion plots were established in the study areas as part of a broader investigation with a four-year duration, this characterization represents the initial phase of the herbivory study.

Keywords:

Dominance, Diversity, Vascular plant communities, Antisana Hydric Conservation Area, Effective Number of Species, Macro-fungal fruiting bodies, White-tailed deer.

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Abbreviations

ACHA: Área de Conservación Hídrica Antisana (Antisana Hydric Conservation Area)

MAE: Ministerio del Ambiente (Ministry of Environment)

FONAG: Fondo para la Protección del Agua (Fund for the Protection of Water)

EPMAPS: Empresa Pública Municipal de Agua Potable y Saneamiento (Municipal Public Company for Drinking Water and Sanitation)

NDVI: Normalized Difference Vegetation Index

GPS: Global Positioning System

LC: Least Concern

NT: Near Threatened

VU: Vulnerable

NMDS: Non-metric Multi-dimensional Scaling

1. Introduction

1.1. Antisana Páramo Botanical History

The more appropriate title for this thesis is “Floristic and Macro-Fungal Characterization in Habitats frequented by White-tailed deer (*Odocoileus virginianus*) in the Paramos of the Antisana Hydric Conservation Area, Ecuador”. This title better described the main objective of this investigation, but due to bureaucracy errors it was not included in the present thesis, this was the reason for this clarification.

The term "páramo," (*sensu lato*), encompasses a collection of high-altitude tropical ecosystems found in South and Central America (Hofstede et al., 2003). Typically characterized by grasslands and a mosaic of bogs and wetlands, these ecosystems are situated in and around mountains and volcanoes, above the tree line, and under perpetual snow (Beltrán et al., 2009; Cuesta et al., 2014). Similar ecosystems exist in Africa and Oceania, albeit under different names (Hofstede et al., 2003; Hofstede et al., 2023).

Páramos in the northern Andes endure significant daily temperature fluctuations, possess nutrient-poor soils, and elevated radiation levels (Ramsay & Oxley, 2001). Despite these challenging conditions, páramos exhibit an exceptionally high vascular flora species richness compared to other high-altitude tropical regions (Sklenář & Jørgensen, 1999; Madriñán et al., 2013).

For over 200 years, the páramo ecosystems of the North Andes were known for their rich plant diversity. Alexander von Humboldt was captivated by the richness of plant species He found in Ecuadorian volcanoes such as Cotopaxi and Antisana. In 1802, Humboldt, Bonpland, and Caldas reunited in Ibarra and visited Quito, where they shared botanical information about the region. Humboldt and Bonpland ascended to the nearest elevations (Gutiérrez, 2016). The majority of plant specimens used for the renowned depictions of mountain vegetation were collected during their attempt to ascend the Antisana Volcano and sole visit to Hacienda

Antisana and Pinantura, thus establishing Antisana volcano as a prominent research site (Madriñán et al., 2013; Grubb et al., 2020).

In his book 'Aspects of Nature,' Humboldt writes, 'No zone of alpine vegetation in the temperate or cold parts of the globe can well be compared with that of the páramos in the tropical Andes.' (Madriñán et al., 2013b, p. 1). The author noted that locals used the word 'páramo' to describe areas of constant cold and rainy weather (Moret et al., 2019, p. 5). However, Humboldt used the terms 'grassland region' for tussock grasses (e.g., *Bromus lanatus* and *Cinnagrostis croacta*) and 'alpine plants region' for cushions and shrubs (e.g., *Azorella pedunculata* and *Chuquiraga jussieu*) instead of 'páramo' in his floristic altitudinal zone classification system of 1805 (Ruales & Guevara, 2010; Gutiérrez, 2016; Moret et al., 2019).

Throughout the 20th and 21st centuries, the traditional definition of páramo ecosystems has evolved from classic altitudinal zonation to regional definitions, taking into account the heterogeneity and asymmetry of zonal belts and plant communities observed in the field (Sklenář & Jørgensen, 1999; Cuesta et al., 2014). Edaphic factors such as gradients in nutrients, humidity, and temperature promote vegetation heterogeneity (Sklenář & Lægaard, 2003), while anthropogenic activities, principally agriculture and burning-grazing activities, have altered vegetation succession and composition (Sklenář & Lægaard, 2003; Cuesta et al., 2014; Peyre & Font, 2015).

Several classifications have been proposed for the north Andes páramos according to the vegetal physiognomy and structure at regional scales (Cuatrecasas A., 1958; Yáñez, P. 1997; Sklenář & Jørgensen, 1999; Beltrán et al., 2009; Cuesta et al., 2017), these authors recognize a typical zonal division into three big groups at the regional level: subpáramo located between approximately 2800 and 3500m, páramo between 3500 and 4200m, and superpáramo between 4200 and 5000 m. The authors also mention the use of typical growth forms of the dominant plants as diagnostic species for some páramos, for example, caulescent rosettes (e.g., *Espeletia pycnophylla*), tussock grasslands (e.g., *Cinnagrostis intermedia*), acaulescent herbs (e.g., *Hypochaeris sessiliflora*), sclerophyllous shrubs (e.g.,

Chuquiraga jussieui) and cushion plants (e.g., *Plantago rigida*). This classification is also described in the Classification System for Ecosystems in Continental Ecuador (MAE, 2012; Hofstede et al., 2023).

Therefore, the dynamic concept of páramo grasslands obtains a definition (*Sensu stricto*) for the north Andean region of Ecuador. Páramo grasslands consist of uneven, mostly glacier-formed valleys with many lakes and are composed of different types of ecosystems intermingled, both zonal and azonal, with shared properties of ultra-humid ombrotypes and cryo-tropical thermotypes (Buytaert et al., 2006; MAE, 2012; Hofstede et al., 2023) with a characteristic zonal abundance of tussock grasslands and patches of azonal caulescent rosettes, cushions, and caulescent herbs.

1.2. Páramo Ecosystem Services

Páramos provide crucial ecosystem services to cities in the North Andean Mountain Range, benefiting not only high-altitude cities such as Quito and Bogotá but also numerous other cities that rely on these services in lower lands (Sklenář & Jørgensen, 1999; Suárez et al., 2013). These high-altitude ecosystems play a critical role in supplying drinking water, irrigation, and hydropower to millions in South America's Andean region (Buytaert et al., 2006). Moreover, páramos are biodiversity hotspots, home to a diverse array of unique and often endemic birds and mammals (Figure 1).

Authors demonstrated that the ability of páramo ecosystems to intercept and store water was heavily influenced by the type of vegetation present (Hofstede et al., 2003; Buytaert et al., 2006; Irazábal-Morales, 2016; FONAG, 2017; Grubb et al., 2020). Certain plant species, such as tussocks and cushion plants, play a significant role in water interception with their foliage and possess deep root systems that penetrate into different soil horizons (Suárez et al., 2013).

Scientific interest in páramo ecosystem services dates back over two centuries. However, evidence suggests indigenous peoples utilized these lands for at least a thousand years before the Spanish conquest of 1588 (Loughlin et al.,

2018). According to the same study, intensive land use for farming occurred before colonization, and indigenous agriculture significantly declined afterward. Subsequently, land use shifted to haciendas for livestock, such as Hacienda Antisana and Pinantura, which Humboldt visited and documented the presence of large herds of cattle and some wild deer, commonly hunted (Grubb et al., 2020).



Figure 1. Iconic fauna in Antisana Hydric Conservation Area (ACHA), Antisana National Park, Ecuador. (A) Andean condor *Vultur gryphus*. (B) The great horned ear owl *Bubo virginianus*. (C) A sequence of Ecuadorian hillstar feeding *Oreotrochilus chimborazo*. (D) Stout-billed cinclodes eating *Cinclodes excelcior* (E) Andean fox *Lycalopex culpaeus*.

As described by Hofstede (1995) in Los Nevados, Colombia, and Velásquez-Romo (2000) in Antisana, Ecuador, burning was a common practice among Andean people for agricultural purposes, primarily to enhance the growth of palatable pastures for livestock. Specifically, Velásquez-Romo (2000) reports intense grazing and trampling pressure within Hacienda Antisana at the site known as "La Ovejería" (FONAG et al., 2022). According to testimony from park rangers, known as "Guardapáramos," who previously worked at Hacienda Antisana, the largest number of livestock, approximately 30,000 animals, mostly sheep, was observed in 2002 (Figure 2). The disturbance to plant cover remains evident, with many areas of exposed soil resulting from heavy herbivory pressure.



Figure 2. Livestock at Ranch Antisana in 2001 (Photo by Francisco Prieto cited by FONAG, 2022). Intensive land use for over two hundred years caused extreme habitat deterioration in large areas.

1.3. Protection of Páramo in Antisana Volcano

One of the primary water sources for Quito City is the Antisana glacier and the surrounding páramos, which serve as natural water catchments (Aguirre et al., 2014). In 1993, the Antisana Ecological Reserve was established to safeguard the glacier and these crucial yet fragile ecosystems. In 2010, public and private funds

were used to buy the Haciendas Antisana and Pinantura (FONAG, 2022) to initiate conservation and restoration projects. One of the initial restoration efforts commenced in 2012, involving the removal of all livestock from the Antisana Ecological Reserve, successfully eliminating sheep and cattle. However, herds of feral horses and llamas still inhabit the Antisana páramos (Grubb et al., 2020).

The Antisana Hydric Conservation Area “Área de Conservación Hídrica Antisana” (ACHA), situated within the former Hacienda Antisana, is managed by the Water Protection Fund "Fondo para la Protección del Agua" (FONAG) and the Public Water Company "Empresa Pública Municipal de Agua Potable y Saneamiento" (EPMAPS) (Aguirre et al., 2014). Conservation practices implemented in ACHA, such as restricted access and hunting prohibition, have facilitated the resurgence of the Andean condor (Figure 1A) and the white-tailed deer (*Odocoileus virginianus*) (Figure 3) (Tellkamp et al., 2019).

The Antisana Ecological Reserve was designed as Antisana Natural Park in 2021 to continue and promote conservation and restoration efforts. Nevertheless, it is threatened by human activities such as urban expansion, road construction, pine plantation, the advance of the agricultural frontier, livestock raising, and mining exploitation (Beltrán et al., 2009; Hofstede et al., 2023). Thompson et al. (2021) say the reduction of the páramos extension in the Antisana and Chimborazo volcanoes is notorious. From 1991 to 2017, the cultivated areas in páramo ecosystems around Quito increased by 838% (Thompson et al., 2021).

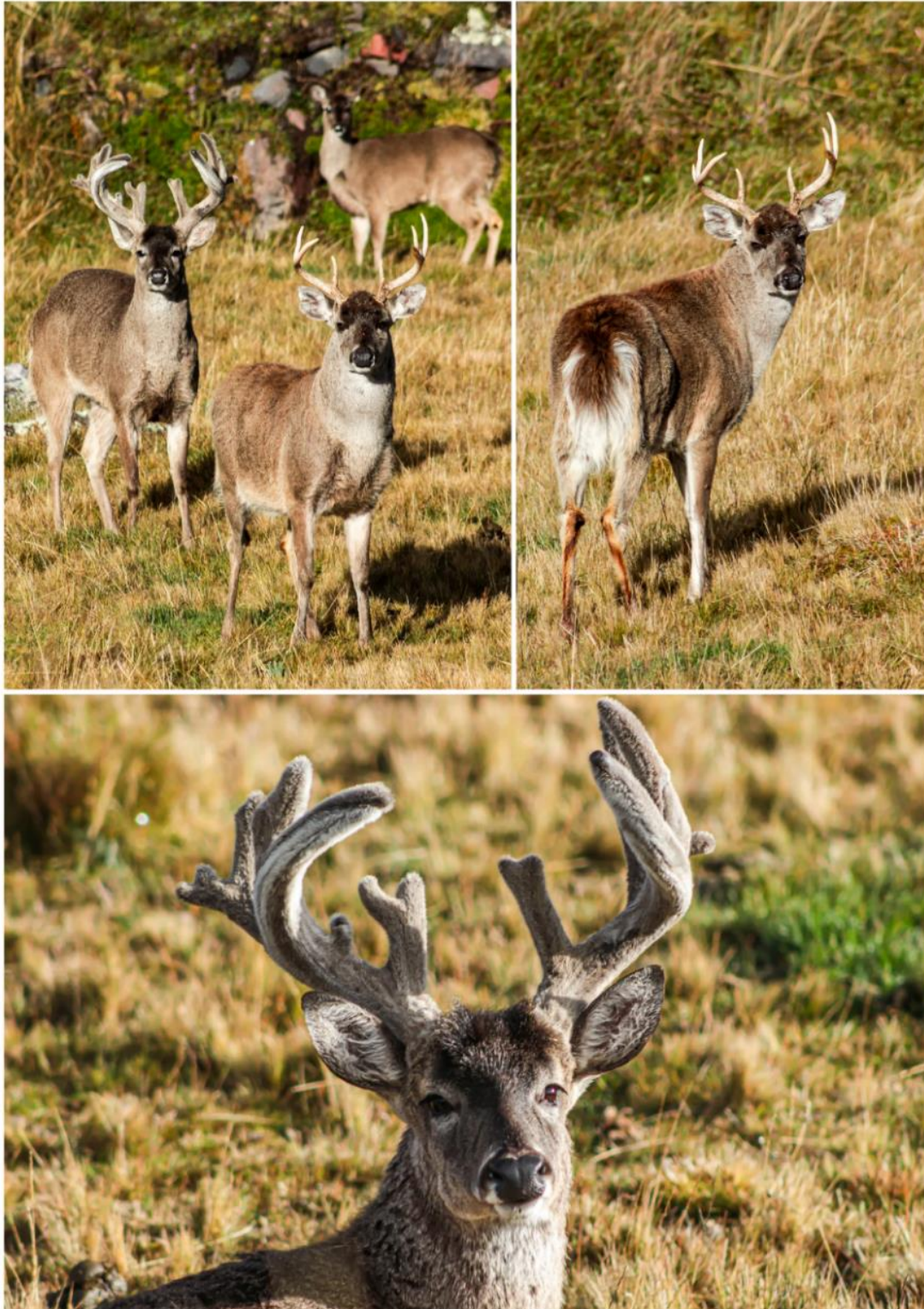


Figure 3. White-tailed deer (*Odocoileus Virginianus*) in ACHA. Herbivory, trampling and browsing can modify plant composition and succession. Also, male specimens can stunt plant growth of bushes by scratching with their antlers to remove the furry skin.

1.4. Plant communities in ACHA

Evidence indicates a difference in plant species composition in the páramo of Antisana compared to other páramos in Ecuador, attributed to the mixture of habitats (Beltrán et al., 2009). Typical páramo landscapes in Ecuador feature extensive areas dominated by tall-tussock species (e.g., *Calamagrostis intermedia*) and shrubby vegetation (e.g., *Chuquiraga jussieui*). However, in the Antisana Hydric Conservation Area (ACHA), large patches of azonal cushion plants (e.g., *Azorella pedunculata* and *Plantago rigida*) and herbaceous vegetation (e.g., *Werneria nubigena* and *Hypochaeris sessiliflora*) are also common. Previous studies on floristic coverage in ACHA have documented differences in floral communities, particularly on overgrazed sites (FONAG, 2017; Cevallos, S, 2022). Grubb et al. (2020), in a historical baseline study of the páramo of Antisana volcano, noted that intense herbivory led to changes in dominance that promoted the introduction of annual grasses at the expense of native and endemic plant species.

1.5. Macrofungal Species in ACHA

Ecological research on páramos has traditionally focused on fauna and flora species, often overlooking the kingdom of fungi (García et al., 2004), despite the evident interactions across all biotic organisms in the ecosystem. For example, Albuja (2007) documented the diet of the white-tailed deer in ACHA, mentioning the fungivory or mycophagy of three mushroom species by the deer. These basidiomycetes were classified as highly palatable to white-tailed deer in ACHA. Evidence suggests that neighboring plants share mycorrhizal networks. These interactions presented positive and negative effects on individual plants and community composition of páramos (Molina-Montenegro, 2015). (Casanova et al., 2011) Thus, they alter plant composition and abundance. Additionally, saprophytic mushrooms degrade organic matter and increase nutrient availability in the soil (García et al., 2004).

2. Objectives

This thesis aims to characterize the richness and biodiversity of flora and report the macrofungal presence in places commonly visited by white-tailed deer in the Antisana Water Conservation Area. The specific objectives of the study are:

1. To describe the taxonomic richness of plant communities at the family and species levels.
2. To report plant communities' dominance patterns (vegetation cover) at the species levels.
3. To assess the diversity indexes and the effective number of species for the selected sites.
4. To assess the community structure of plants in ACHA using a NMDS.
5. To describe macroscopic fungi growing inside the study sites.

These specific objectives are descriptive and require no hypotheses to be proven.

It is crucial to note that this study marks the beginning of a longer research project on herbivory. This report is a visualization of the floristic composition of the plots at time zero, which will serve as the basis for implementing exclusion treatments for a 4-year study about herbivory.

3. Materials and Methods

3.1. Study Area

The study area, the Antisana Hydric Conservation Area (Figure 4), is situated within Antisana National Park on the western slope of the Antisana volcano, approximately 70 km east of Quito, spanning the border of Napo and Pichincha provinces. Covering an area of 94.45 km², it boasts an altitudinal range of 3720-4760m above sea level, with local ecosystems classified as páramo (3500-4200m) and superpáramo (4200-5000m) (MAE, 2012; Yáñez, 2014; Hofstede et al., 2023). The annual precipitation is approximately 1400 mm, with April recording the highest precipitation levels (Lahuatte et al., 2015).

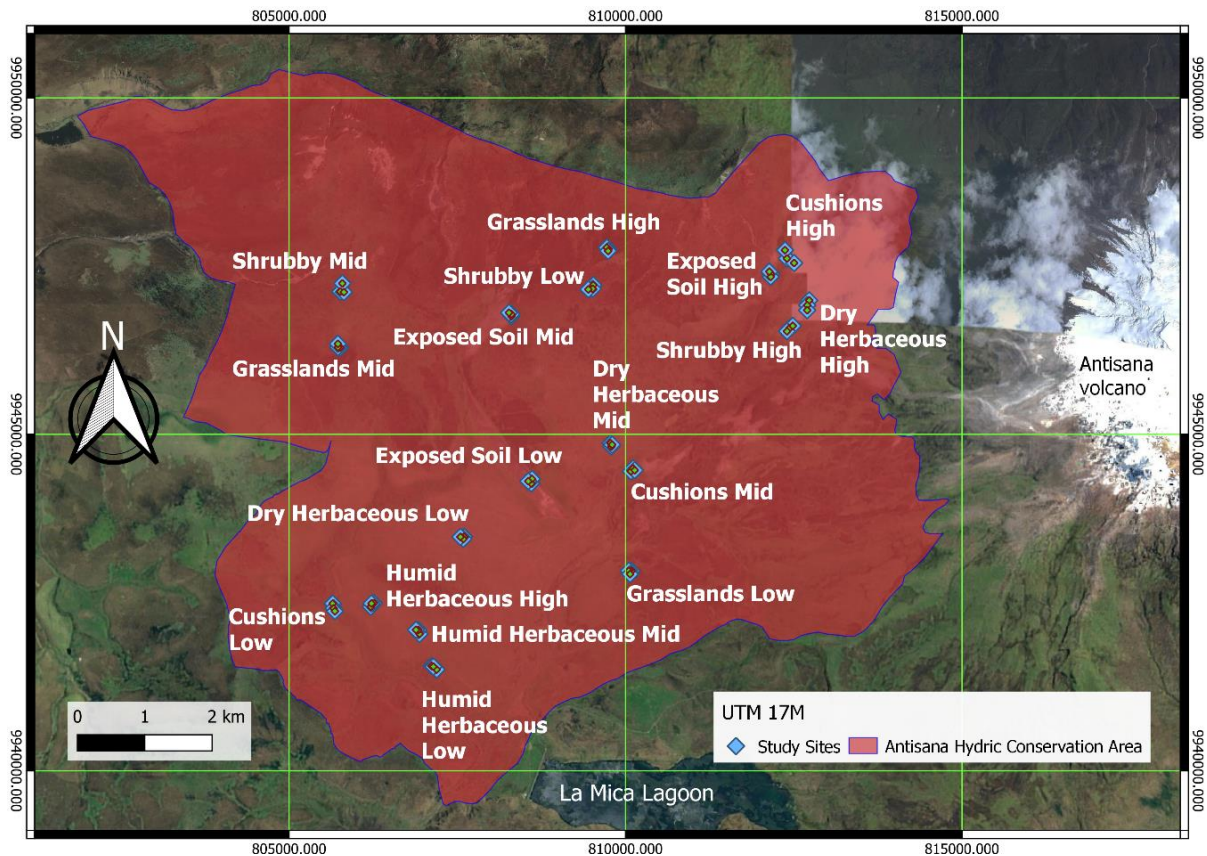


Figure 4. The Antisana Hydric Conservation Area (red polygon) is located in the western downslopes of the Antisana volcano. Marked in blue are the 54 plots distributed on the different habitat and altitudinal categories labeled in white.

The Antisana Hydric Conservation Area (ACHA) was selected as the study area for this research due to the vital role of vegetative cover in water catchment. Additionally, the rising population density of white-tailed deer in the area (Tellkamp et al., 2019) poses a potential threat to vegetative cover.

Recently, Tellkamp et al. (2019) identified six habitat categories within ACHA that are significant for the utilization and distribution of deer. This study incorporates the categories proposed by Tellkamp et al. (2019) to delineate the study sites. This classification reflected the heterogeneity of local ecosystems and was developed based on satellite imagery, particularly utilizing the Normalized Difference Vegetation Index in QGIS software. The classification of these habitat categories includes diagnostic plant species and geomorphological factors.

1. **Grassland vegetation** with abundant tall tussock grasses (characteristic species: *Cinnagrostis intermedia* and *Stellaria recurvata*).
2. **Humid Herbaceous vegetation** with occasional cushion plants (*Bromus lanatus* and *Plantago rigida*).
3. **Dry Herbaceous vegetation** presents a similar composition to the previous habitat classification but higher photosynthesis according to NDVI (*Cinnagrostis croacta* and *Valeriana rigida*).
4. **Cushion plants** in humid valley bottoms near water catchments (*Plantago rigida* and *Distichia muscoides*).
5. **Shrubby vegetation** with a mixture of herbs and cushion plants (*Chuquiraga jussieui* and *Lasiocephalus ovatus*).
6. **Exposed soil** as bare ground with scarce vegetation (*Hypochaeris sessiliflora* and *Lupinus microphyllus*).

The plots were placed systematically in randomly selected places. All the parameters for the placement of the plots were the altitudinal gradient, presence of characteristic species, avoidance of big slopes to prevent seed rain and shadow, and at least 50 meters away from roads, rivers, and habitat margins. Additionally, each plot was separated by a distance of 100 meters from each other.

Each plot is labeled with an abbreviation representing a combination of three words: the habitat category name, an altitudinal category (Low, Mid, High), and the type of exclusion treatment (Control, Deer exclusion, Rabbit exclusion). These exclusion treatments are designed to demonstrate the effects of herbivory absence over time. Therefore, in this initial characterization, the exclusion types are used solely as names for the plots (Table 1).

Table 1. The name of each plot consisted of the habitat and altitudinal categories combined with the type of exclusion treatment; the label for the plot is the abbreviation of its name.

Habitat categories	Altitudinal categories within each habitat		
	Low	Mid	High
Cushion plants	1 Cushion Low Control (CLC) Plot	1 Cushion Mid Control (CMC) Plot	1 Cushion High Control (CHC) Plot
	1 Cushion Low Deer (CLD) Exclusion Plot	1 Cushion Mid Deer (CMD) Exclusion Plot	1 Cushion High Deer (CHD) Exclusion Plot
	1 Cushion Low Rabbit (CLR) Exclusion Plot	1 Cushion Mid Rabbit (CMR) Exclusion Plot	1 Cushion High Rabbit (CHR) Exclusion Plot
Shrubby vegetation	1 Shrubby Low Control (SLC) Plot	1 Shrubby Mid Control (SMC) Plot	1 Shrubby High Control (SHC) Plot
	1 Shrubby Low Deer (SLD) Plot	1 Shrubby Mid Deer (SMD) Exclusion Plot	1 Shrubby High Deer (SHD) Exclusion Plot
	1 Shrubby Low Rabbit (SLR) Plot	1 Shrubby Mid Rabbit (SMR) Exclusion Plot	1 Shrubby High Rabbit (SHR) Exclusion Plot
Dry Herbaceous vegetation	1 Dry Herbaceous Low Control (DLC) Plot	1 Dry Herbaceous Mid Control (DMC) Plot	1 Dry Herbaceous High Control (DHC) Plot
	1 Dry Herbaceous Low Deer (DLD) Exclusion Plot	1 Dry Herbaceous Mid Deer (DMD) Exclusion Plot	1 Dry Herbaceous High Deer (DHD) Exclusion Plot
	1 Dry Herbaceous Low Rabbit (DLR) Exclusion Plot	1 Dry Herbaceous Mid Rabbit (DMR) Exclusion Plot	1 Dry Herbaceous High Rabbit (DHR) Exclusion Plot

Habitat categories	Altitudinal categories within each habitat		
	Low	Mid	High
Humid Herbaceous vegetation	1 Humid Herbaceous Low Control (HLC) Plot	1 Humid Herbaceous Mid Control (HMC) Plot	1 Humid Herbaceous High Control (HHC) Plot
	1 Humid Herbaceous Low Deer (HLD) Exclusion Plot	1 Humid Herbaceous Mid Deer (HMD) Exclusion Plot	1 Humid Herbaceous High Deer (HHD) Exclusion Plot
	1 Humid Herbaceous Low Rabbit (HLR) Exclusion Plot	1 Humid Herbaceous Mid Rabbit (HMR) Exclusion Plot	1 Humid Herbaceous High Rabbit (HHR) Exclusion Plot
Exposed soil	1 Exposed Soil Low Control (ELC) Plot	1 Exposed Soil Mid Control (EMC) Plot	1 Exposed Soil High Control (EHC) Plot
	1 Exposed Soil Low Deer (ELD) Exclusion Plot	1 Exposed Soil Mid Deer (EMD) Exclusion Plot	1 Exposed Soil High Deer (EHD) Exclusion Plot
	1 Exposed Soil Low Rabbit (ELR) Exclusion Plot	1 Exposed Soil Mid Rabbit (EMR) Exclusion Plot	1 Exposed Soil High Rabbit (EHR) Exclusion Plot
Grassland	1 Grassland Low Control (GLC) Plot	1 Grassland Mid Control (GMC) Plot	1 Grassland High Control (GHC) Plot
	1 Grassland Low Deer (GLD) Exclusion Plot	1 Grassland Mid Deer (GMD) Exclusion Plot	1 Grassland High Deer (GHD) Exclusion Plot
	1 Grassland Low Rabbit (GLR) Exclusion Plot	1 Grassland Mid Rabbit (GMR) Exclusion Plot	1 Grassland High Rabbit (GHR) Exclusion Plot

All 54 plots were positioned following a close examination of the sites by two academic experts and one guardapáramo. The locations were identified, and their coordinates were recorded using a global positioning system (GPS) (see Annex 1).

Details regarding the construction process and characteristics of the exclusions are provided in the annex section (see Annex 2). Fieldwork in ACHA was conducted between April 2022 and July 2023, spanning 40 effective workdays.

3.2. Sampling Vascular Plants

Species abundance and dominance were estimated by measuring vegetation cover using the point-intercept method, known for its speed and efficiency in low vegetation areas (Floyd & Anderson, 1987). This method is primarily suited for vegetation less than 1m in height and is highly sensitive for measuring ground cover (Caratti, 2006; Cuesta et al., 2014; FONAG, 2022). In this approach, multiple equidistant points were separated by 20 cm, recording the presence or absence of local species. A straight rod was placed perpendicular to the ground at each point. A hit was recorded only when one or more plant species directly touched the rod; otherwise, it was marked as bare ground. A total of 96 points were conducted in each plot (Figure 5). During the application of the methodology (Figure 6), it was crucial to consider contact with the rod at different levels from the ground to the top.

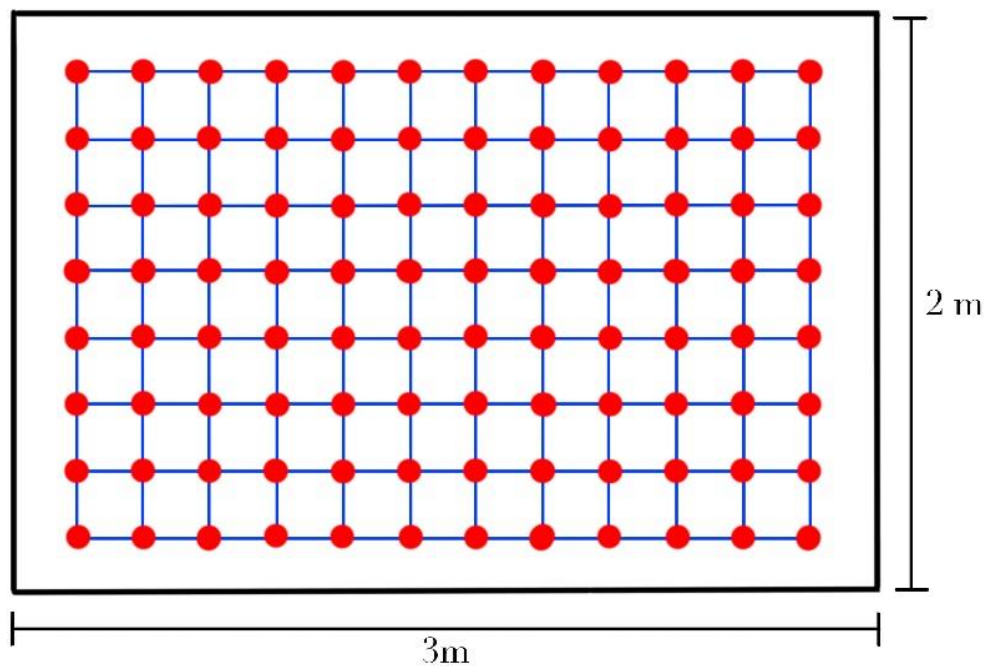


Figure 5. Diagram of the point-intercept method for calculating plant cover, with 96 equidistant points in 12 columns with 8 rows. All sites have a 3m length and 2m width. The points were equidistant at 20 cm from each other.



Figure 6. Application of the point-intercept methodology for estimating the vegetation cover in the pots at Antisana Hydric Conservation Area (ACHA), Antisana National Park, Ecuador.

3.3. Taxonomic identification

Botanical samples were collected from outside the plots using conventional methods. Each voucher was photographed and promptly placed in a sealed plastic bag. At least one voucher was collected per species, with a maximum of three duplicates from the same individual. Each botanical voucher was labeled with a corresponding code in the database. Subsequently, the samples were pressed and dried.

Taxonomic identification of each specimen was conducted using a lens, stereoscope, systematic books, páramo flora field guides, internet checklists (such as The Catalogue of Vascular Plants of Ecuador 2023 in Tropicos and World Flora Online 2023), and expert opinions (Marcia Peñafiel, Patricio Yanez). All taxonomical data were verified at the National Herbarium of Ecuador (INABIOEC-QCNE) for a final determination.

3.4. Macrofungal Identification and Description

Only specimens growing directly inside the plots during the floristic characterization were considered. The diameter of each mushroom was measured, and the approximate mushroom area was calculated. Specimens were collected for identification using specialized guides, such as the photographic guide "Hóngos del páramo" from The Fungi Web Initiative (Ordoñez, 2018). Morphological characters were described using a stereoscope. The mycological vouchers were placed in labeled paper bags and dried following standard protocols. Spores were photographed under 1000x magnification using an optical microscope.

3.5. Data Analyses

3.5.1. Percent Cover of Plant Species

Data were recorded in several spreadsheets. Subsequently, they were organized, analyzed, and plotted using R version 4.3.1 in RStudio (2023) with various packages (Annex 3).

The percentage of cover by each species was calculated using a simple equation (Equation 1).

$$\% \text{ Cover of species } j = \frac{\text{Number of hits in contact with species } j}{\text{Total number of hits done per plot (96)}} \times (100)$$

Equation 1. Percentage of vegetation cover for one single species.

Dominance was evaluated at the species level; for this, the percentage cover of an individual species divided by the total percentage cover of all species was calculated (Equation 2).

$$P_{ij} = \frac{\% \text{ cover of species } j}{\text{Total } \% \text{ cover of all species in the site}}$$

Equation 2. Proportion of plant cover of a single species (j) in relation to the total plant cover of all species in the site.

3.5.2. Rank-Abundance Plots or Whittaker Curves

To elaborate a rank- abundance curve or Whittaker curve (Yáñez, 2014), the values of P_i were ordered in descending order for each site. Then, the values were plotted with the sequence of species on the Y-axis and the corresponding value of P_i on the X-axis. The Y-axis was displayed in a logarithmic fashion. In this manner, species with a P_i value from 1 to 0.1 could be designated as dominant, from 0.1 to 0.01 as co-dominant, and from 0.01 to 0.001 as rare species.

3.5.3. Diversity Indexes

Shannon Diversity Index was calculated for every site using the following formula (Equation 3) (Magurran, 2004):

$$H' = - \sum_{i=1}^S (P_i)(\log_{\text{nat}} P_i)$$

Equation 3. Shannon Diversity Index, where H' = Shannon Diversity Index; S = Number of species; P_i = proportion of one species plant cover in the sample related to the total plant cover in the sample.

Also, the Simpson Diversity Index was calculated considering the next formula, being (Equation 4):

$$D = 1 - \sum_{i=1}^S P_i^2$$

Equation 4. Simpson Diversity Index. Where: D = Simpson Diversity Index; S = Number of species; P_i = proportion of one species plant cover in the sample related to the total plant cover.

3.5.4. Effective Number of Species

The nonlinearity of diversity indices can lead to errors if compared directly (Jost, 2006). So, to compare the diversity of different plots, the diversity indices were converted to true diversity or the Effective Number of Species (ENS).

The Effective Number of Species based on the Shannon Diversity Index was calculated using the following formula (Equation 5).

$$ENS(H') = \exp(H')$$

Equation 5. The Effective Number of Species based on the Shannon Diversity Index. Where: H' = Shannon Diversity Index.

The Effective Number of Species based on the Simpson Diversity Index was calculated using the following formula (Equation 6).

$$ENS(D) = \frac{1}{1 - D}$$

Equation 6. The Effective Number of Species based on the Simpson Diversity Index. Where: D = Simpson Diversity Index.

For a plot with dominant species, the Effective Number of Species based on the Shannon Diversity Index is always less than the species richness, and the Effective Number of Species based on the Simpson Diversity Index is always less than the Effective Number of Species based on the Shannon Diversity Index. The Effective Number of Species is the number of equally abundant species necessary to produce the observed value of diversity (Jost, 2019).

4. Results

4.1. Plant Characterization

4.1.1. Taxonomic richness

In all 54 plots of this study (each 6 m²), I identified 23 families of tracheophytes, encompassing 47 genera and 69 species (Figure 7). Among these, twelve species were endemic. Notably, three of the endemic species held a "Vulnerable" conservation status: *Halenia minima* (Gentianaceae), *Gentianella rupicola* (Gentianaceae), and *Geranium sericeum* (Geraniaceae). Additionally, two endemic species were classified as "Near Threatened" in terms of conservation status: *Festuca chimborasensis* (Poaceae), and *Draba obovata* (Brassicaceae).

Five were classified with "Least Concern" conservation status: *Aphanactis ollgaardii* (Asteraceae), *Astragalus geminiflorus* (Fabaceae), *Carex toreadora* (Cyperaceae), *Castilleja nubigena* (Scrophulariaceae), and *Distichia acicularis* (Juncaceae) (León-Yáñez et al. 2011; IUCN, 2020). Furthermore, two endemic species lacked information regarding their conservation status: *Stachys grandidentata* (Lamiaceae), and *Stellaria recurvata* (Caryophyllaceae).

The remaining 56 species were native and did not possess a threatened conservation status. Additionally, one introduced species was present: *Taraxacum officinale* (Asteraceae) (Annex 3).

The most speciose tracheophyte family was Asteraceae, with 16 species. The second most speciose was Poaceae with eight species, and third, Apiaceae with five species. The families Scrophulariaceae, Geraniaceae, Gentianaceae, and Caryophyllaceae each had 4 species. The families Plantaginaceae and Cyperaceae each had three species. The families Rosaceae, Ranunculaceae, and Juncaceae each had two species. The remaining families, including Violaceae, Valerianaceae, Ophioglossaceae, Montiaceae, Malvaceae, Lycopodiaceae, Lamiaceae,

Ephedraceae, Campanulaceae, and Brassicaceae, each had only one species (Figure 7).

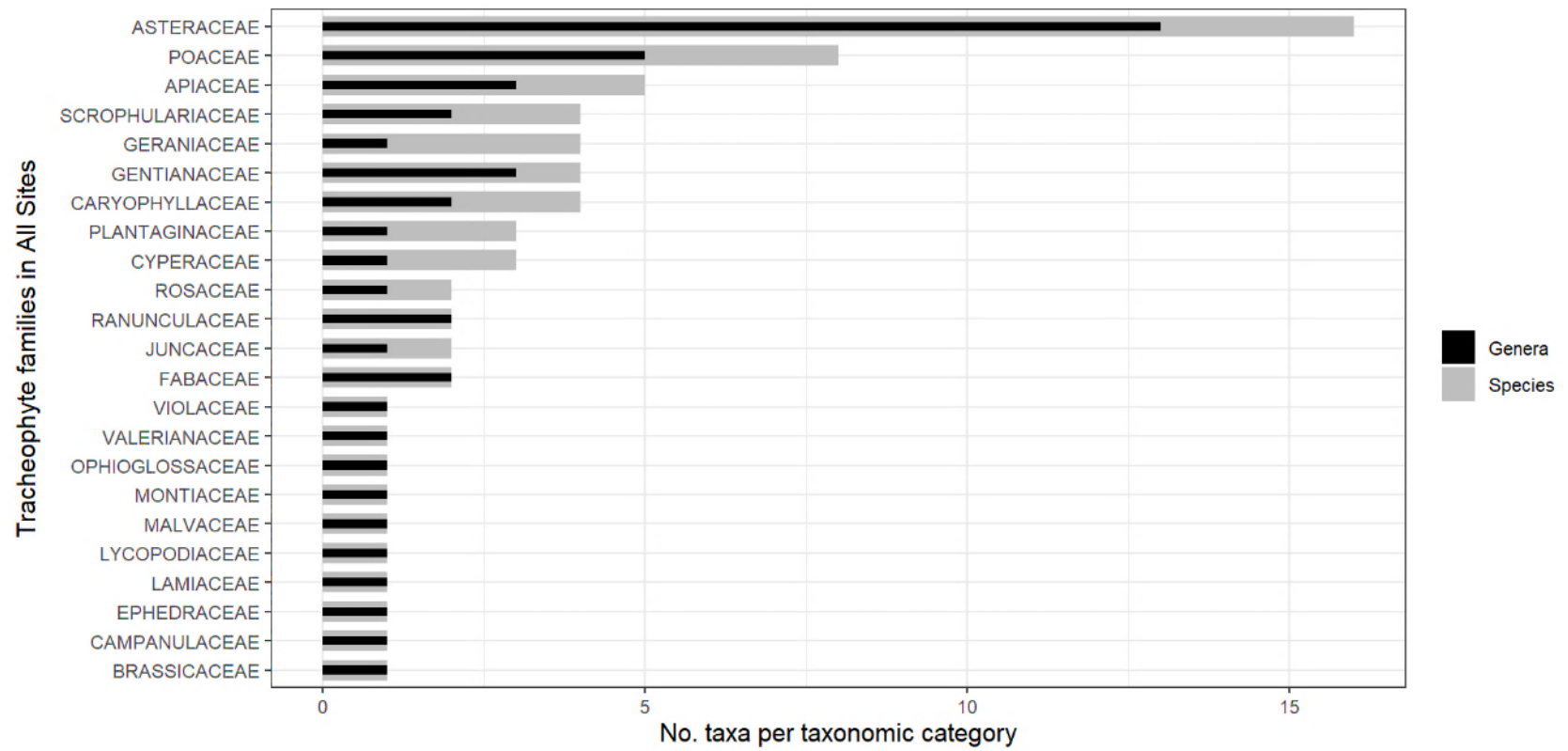


Figure 7. The number of families, genera, and species found at Antisana Hydric Conservation Area (ACHA) study sites in Antisana National Park, Ecuador.

The five most species-rich plots were: “Shrubby High Rabbit” with 23 species, “Dry Herbaceous High Deer” with 22 species, “Shrubby High Control” with 20 species, “Dry Herbaceous Low Rabbit” with 18 species, “Shrubby High Deer” with 18 species. The five least species-rich were: “Exposed Soil Mid Deer” with 6, “Grasslands Mid Control” with 7, “Exposed Soil Mid Rabbit” with 8 species, “Grasslands Low Control” with 9, and “Exposed Soil Mid Control” with 9 species (Figure 8).

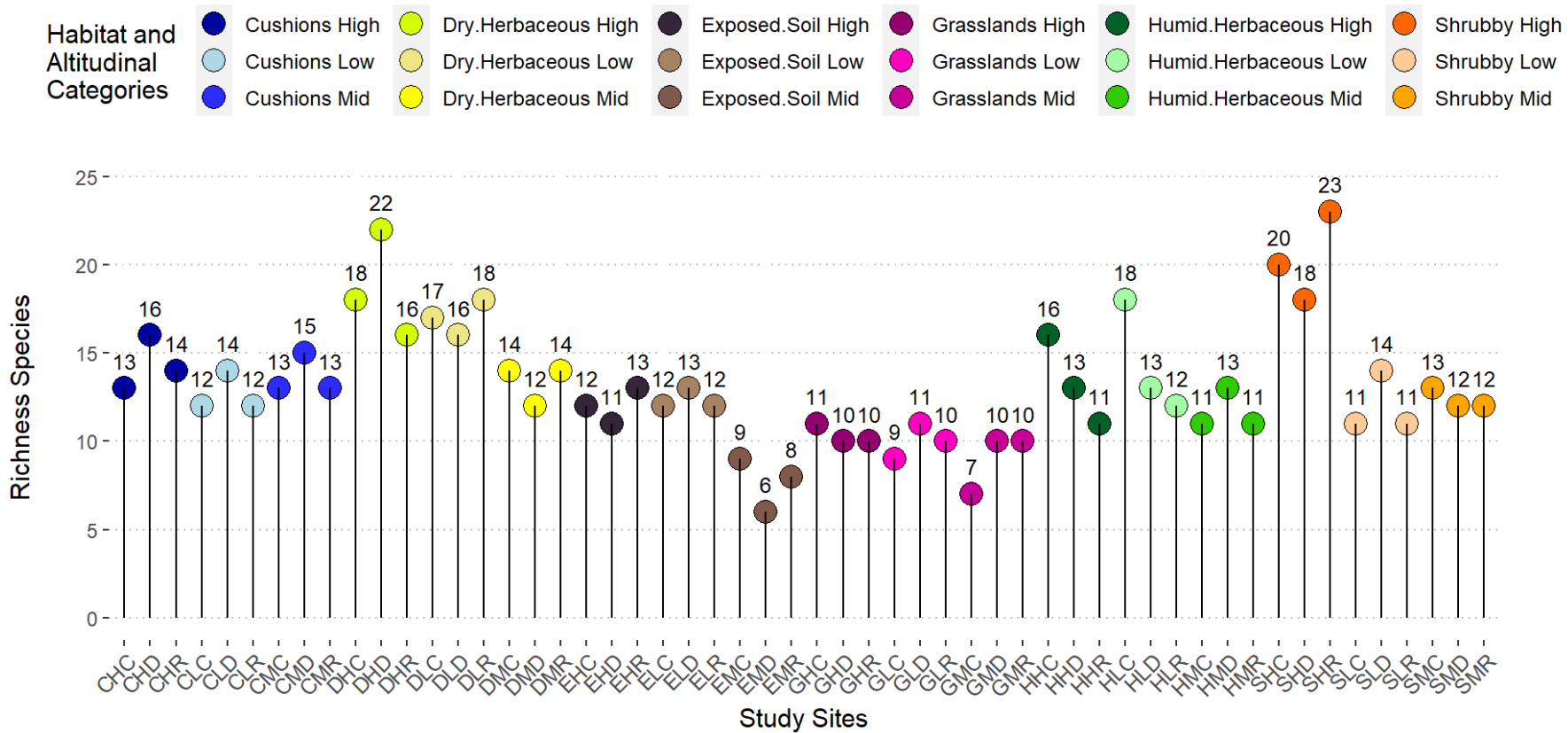


Figure 8. The richness of vascular plant species at the Antisana Hydric Conservation Area (ACHA) study sites, Antisana National Park, Ecuador.

4.1.2. Diversity Indices

The five most diverse plots according to the Shannon index (Figure 9) were “Shrubby High Rabbit” ($H' = 2,64$), “Dry Herbaceous Low Rabbit” ($H' = 2,59$), “Dry Herbaceous Low Control” ($H' = 2,54$), “Shrubby High Deer” ($H' = 2,45$), “Dry Herbaceous High Deer” ($H' = 2,42$). Conversely, the five least diverse were “Grasslands High Rabbit” ($H' = 0,99$), “Grasslands Low Control” ($H' = 1,04$), “Grasslands High Control” ($H' = 1,04$), “Grasslands Mid Control” ($H' = 1,06$), “Grasslands Mid Deer” ($H' = 1,07$) (Table 2).

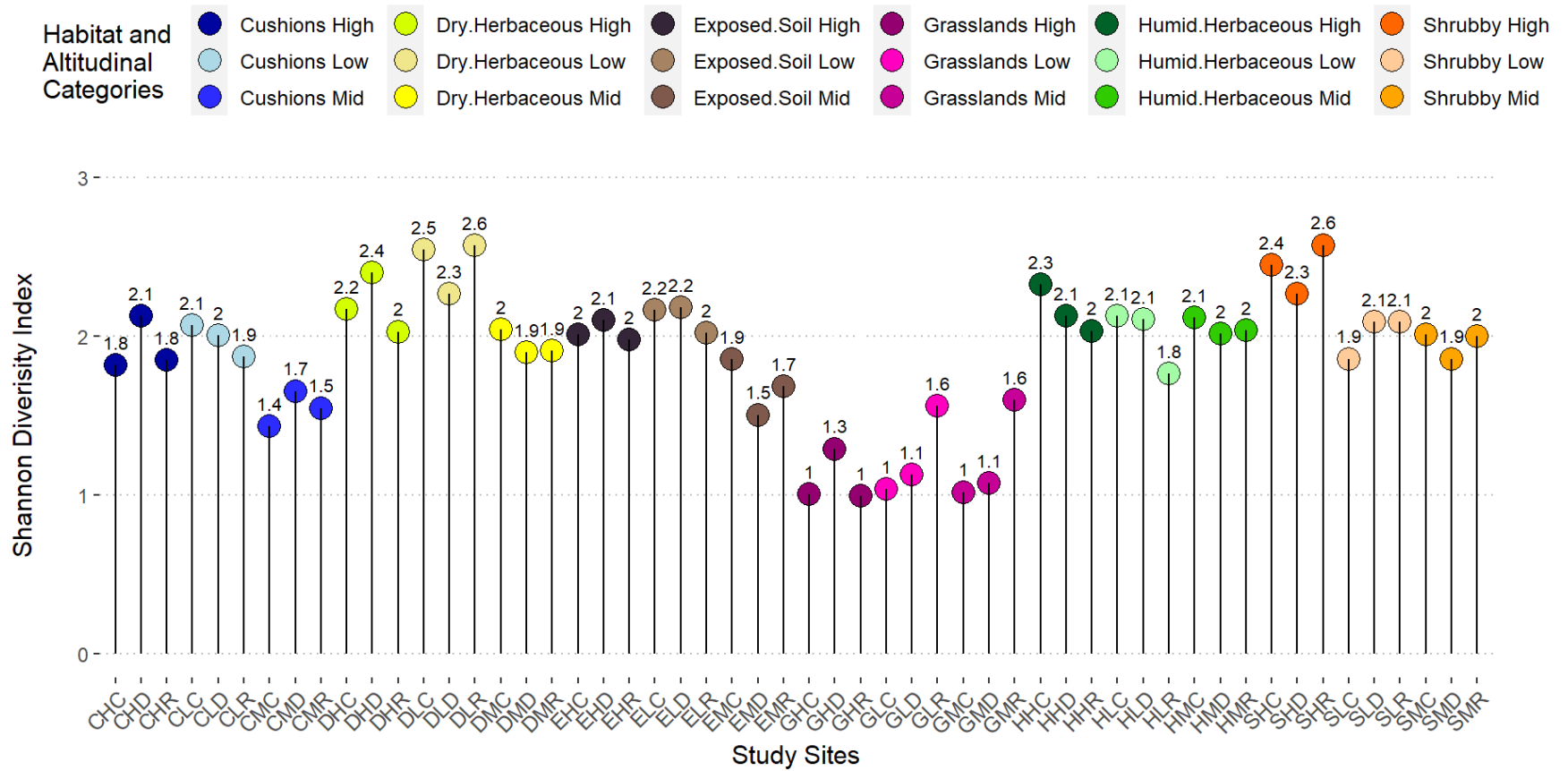


Figure 9. Shannon Diversity Index at the Antisana Hydric Conservation Area (ACHA) study sites, Antisana National Park, Ecuador.

The five most diverse plots according to the Simpson Diversity Index (Figure 10) were “Dry Herbaceous Low Control” ($D = 0,90$), “Dry Herbaceous Low Rabbit” ($D = 0,90$), “Shrubby High Rabbit” ($D = 0,90$), “Humid Herbaceous High Control” ($D = 0,89$), “Shrubby High Control” ($D = 0,89$). Conversely, the five least diverse were “Grasslands High Control” ($D = 0,394$), “Grasslands High Rabbit” ($D = 0,42$), “Grasslands Low Control” ($D = 0,44$), “Grasslands Mid Deer” ($D = 0,46$), and “Grasslands Mid Control” ($D = 0,48$) (Table 2).

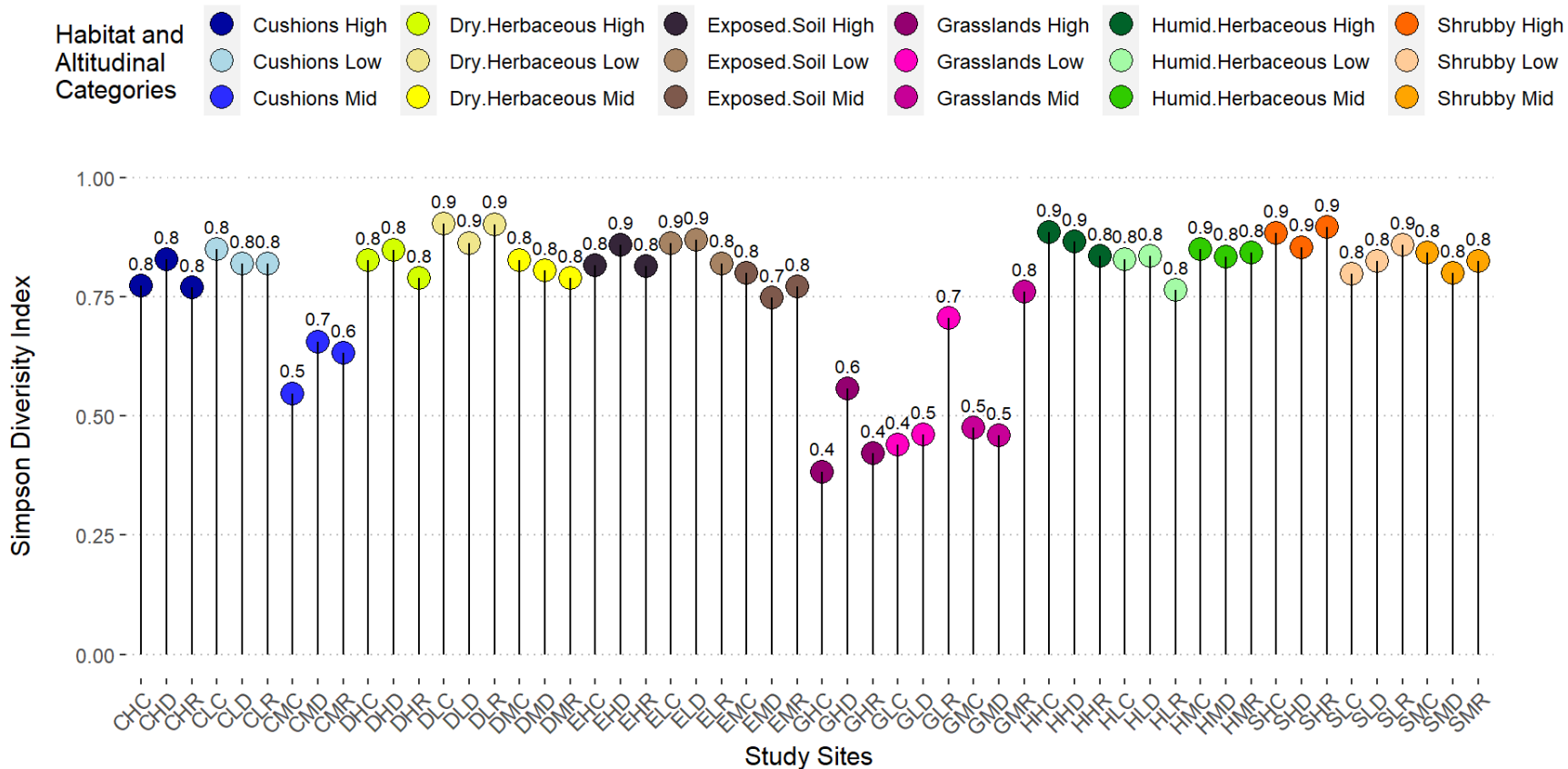


Figure 10. Simpson Diversity Index was found at the Antisana Hydric Conservation Area (ACHA) study sites, Antisana National Park, Ecuador.

4.1.3. Effective Number of Species

The five most diverse plots, according to the Effective Number of Species based on the Shannon Diversity Index (Figure 11), were “Shrubby High Rabbit” (13 effective spp.), “Dry Herbaceous Low Rabbit” (13 effective spp.), “Dry Herbaceous Low Control” (13 effective spp.); “Shrubby High Control” (12 effective spp.); and “Dry Herbaceous High Deer” (11 effective spp.). Conversely, the least diverse plots according to the same Index were “Grasslands High Rabbit” (3 effective spp.), “Grasslands High Control” (3 effective spp.), “Grasslands Mid Control” (3 effective spp.), “Grasslands Low Control” (3 effective spp.), “Grasslands Mid Deer” (3 effective spp.) (Table 2).

Table 2. Effective Number of Species, Shannon Diversity Index, Simpson Diversity Index, Richness, Effective Number of Species based on the Shannon Diversity Index, and Effective Number of Species based on the Simpson Diversity Index.

Name of the Site	H'	D	Richness	ENS(H')	ENS(D)
Cushions High Control	1,81	0,77	13	6	4
Cushions High Deer	2,13	0,83	16	8	6
Cushions High Rabbit	1,85	0,77	14	6	4
Cushions Low Control	2,07	0,85	12	8	7
Cushions Low Deer	2,01	0,82	14	7	6
Cushions Low Rabbit	1,87	0,82	12	6	6
Cushions Mid Control	1,43	0,55	13	4	2
Cushions Mid Deer	1,65	0,66	15	5	3
Cushions Mid Rabbit	1,55	0,63	13	5	3
Dry Herbaceous High Control	2,17	0,83	18	9	6
Dry Herbaceous High Deer	2,40	0,85	22	11	7
Dry Herbaceous High Rabbit	2,03	0,79	16	8	5
Dry Herbaceous Low Control	2,54	0,90	17	13	10
Dry Herbaceous Low Deer	2,27	0,86	16	10	7
Dry Herbaceous Low Rabbit	2,57	0,90	18	13	10
Dry Herbaceous Mid Control	2,04	0,83	14	8	6
Dry Herbaceous Mid Deer	1,90	0,81	12	7	5
Dry Herbaceous Mid Rabbit	1,91	0,79	14	7	5
Exposed Soil High Control	2,01	0,82	12	7	5
Exposed Soil High Deer	2,10	0,86	11	8	7
Exposed Soil High Rabbit	1,98	0,81	13	7	5

Name of the Site	H'	D	Richness	ENS(H')	ENS(D)
Exposed Soil Low Control	2,16	0,86	12	9	7
Exposed Soil Low Deer	2,18	0,87	13	9	8
Exposed Soil Low Rabbit	2,02	0,82	12	8	6
Exposed Soil Mid Control	1,85	0,80	9	6	5
Exposed Soil Mid Deer	1,50	0,75	6	4	4
Exposed Soil Mid Rabbit	1,68	0,77	8	5	4
Grasslands High Control	1,00	0,38	11	3	2
Grasslands High Deer	1,29	0,56	10	4	2
Grasslands High Rabbit	0,99	0,42	10	3	2
Grasslands Low Control	1,04	0,44	9	3	2
Grasslands Low Deer	1,13	0,46	11	3	2
Grasslands Low Rabbit	1,56	0,70	10	5	3
Grasslands Mid Control	1,02	0,48	7	3	2
Grasslands Mid Deer	1,07	0,46	10	3	2
Grasslands Mid Rabbit	1,60	0,76	10	5	4
Humid Herbaceous High Control	2,32	0,89	16	10	9
Humid Herbaceous High Deer	2,13	0,87	13	8	7
Humid Herbaceous High Rabbit	2,03	0,84	11	8	6
Humid Herbaceous Low Control	2,13	0,83	18	8	6
Humid Herbaceous Low Deer	2,11	0,84	13	8	6
Humid Herbaceous Low Rabbit	1,76	0,76	12	6	4
Humid Herbaceous Mid Control	2,12	0,85	11	8	7
Humid Herbaceous Mid Deer	2,01	0,83	13	7	6
Humid Herbaceous Mid Rabbit	2,04	0,84	11	8	6
Shrubby High Control	2,45	0,88	20	12	9
Shrubby High Deer	2,26	0,85	18	10	7
Shrubby High Rabbit	2,57	0,90	23	13	10
Shrubby Low Control	1,86	0,80	11	6	5
Shrubby Low Deer	2,09	0,82	14	8	6
Shrubby Low Rabbit	2,09	0,86	11	8	7
Shrubby Mid Control	2,01	0,84	13	7	6
Shrubby Mid Deer	1,86	0,80	12	6	5
Shrubby Mid Rabbit	2,00	0,82	12	7	6

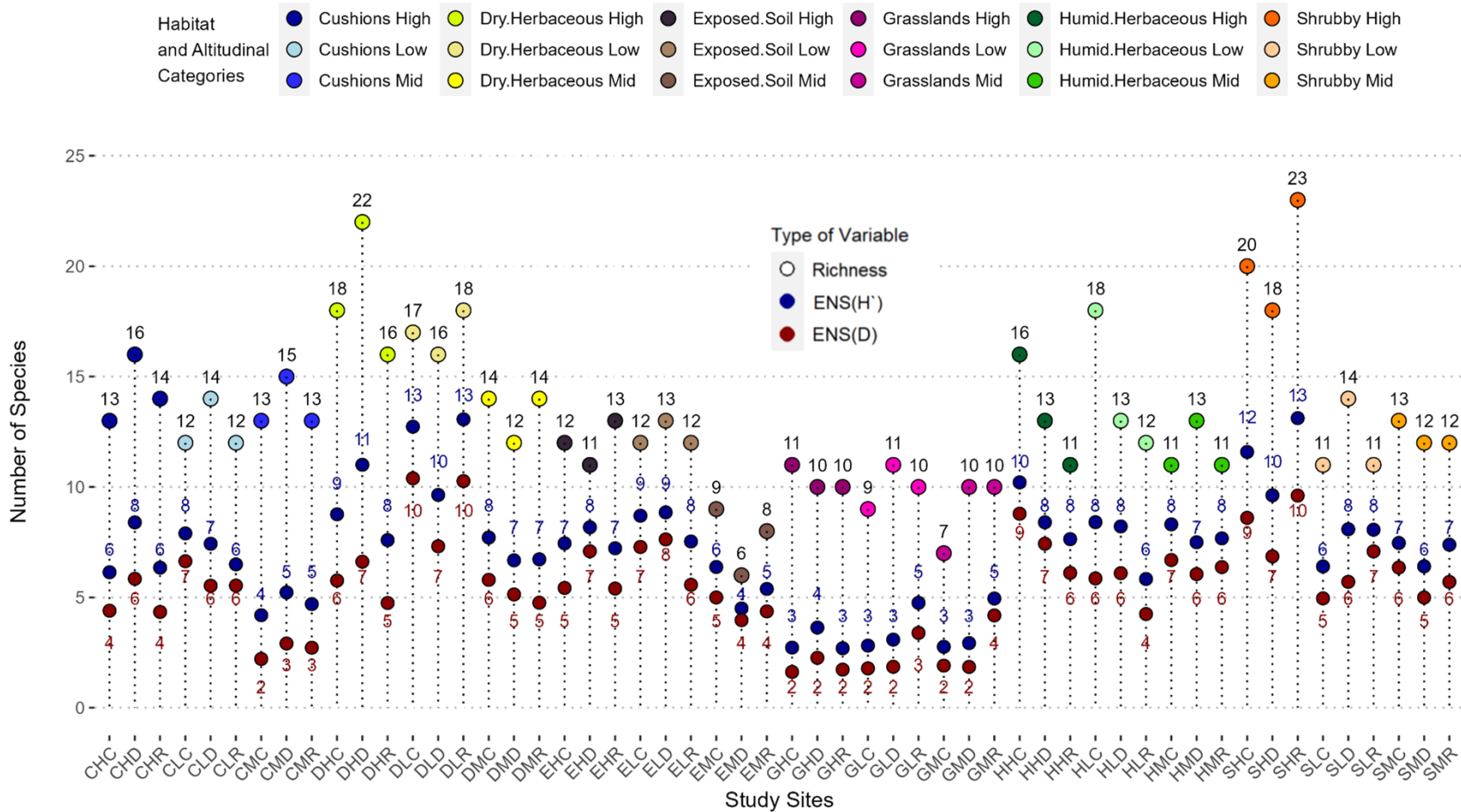


Figure 11. Richness, Effective Number of Species based on Shannon and Simpson diversity Indexes for all plots at the Antisana Hydric Conservation Area (ACHA), Antisana National Park, Ecuador.

4.1.4. Rank Abundance Curves

The Whittaker curves for the three plots in each Habitat and Altitudinal category were displayed in a single graph to analyze the abundance of plant species.

The three most dominant species in the Cushion plants habitat category were *Plantago rigida* (Plantaginaceae), *Azorella pedunculata* (Apiaceae), and *Alchemilla propinqua* (Rosaceae). Numerous co-dominant species were also observed, including *Huperzia crassa* (Lycopodiaceae), and *Baccharis caepitosa* (Asteraceae). The three most rare species were *Halenia minima* (Gentianaceae), *Lysipomia montioides* (Campanulaceae), and *Neobartsia pedicularoides* (Scrophulariaceae) (Figure 12).

Overall, the “Cushions Low” (Figure 12A) and “Cushions High” (Figure 12C) habitat and altitudinal categories exhibited at least three dominant species, whereas the “Cushions Mid” (Figure 12B) category plots only demonstrated one or two dominant species. Across all the plots, there was a high number of co-dominant species. At the same time, every plot had at least two or three rare plant species.

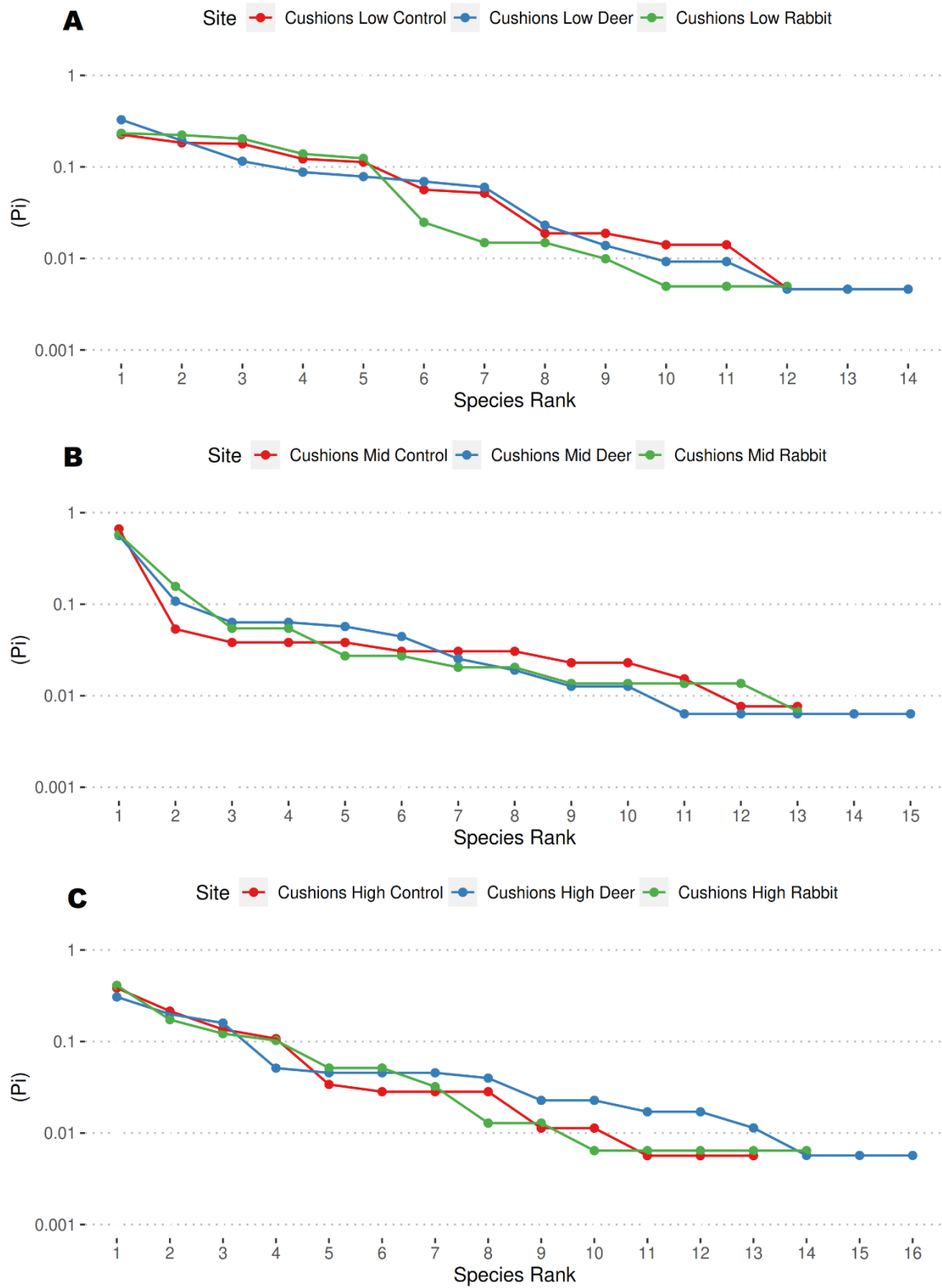


Figure 12. Whittaker curves for all sites in the “Cushion Plants” habitat category. (A) “Low” altitudinal category. (B) “Mid” altitudinal category. (C) “High” altitudinal category.

In the case of the "Dry Herbaceous" habitat category plots, the three most dominant plants were *Werneria nubigena* (Asteraceae), *Valeriana rigida* (Valerianaceae) and *Geranium multipartitum* (Geraniaceae). Many codominant species included *Xenophyllum humile* (Asteraceae); and *Festuca chimbrazensis* (Poaceae). The three most rare species were *Senecio chionogeton* (Asteraceae), *Viola bangii* (Violaceae), and *Werneria pygmaea* (Asteraceae) (Figure 13).

Two of the sites exhibited similar behavior, namely "Dry Herbaceous Low" (Figure 13A) and "Dry Herbaceous Mid" (Figure 13B), with two or three dominant plants, many co-dominant plant species, and at least one rare species. The last category, "Dry Herbaceous High" (Figure 13C), presented different behavior, with at least four rare plant species in the plots and two or three dominant species.

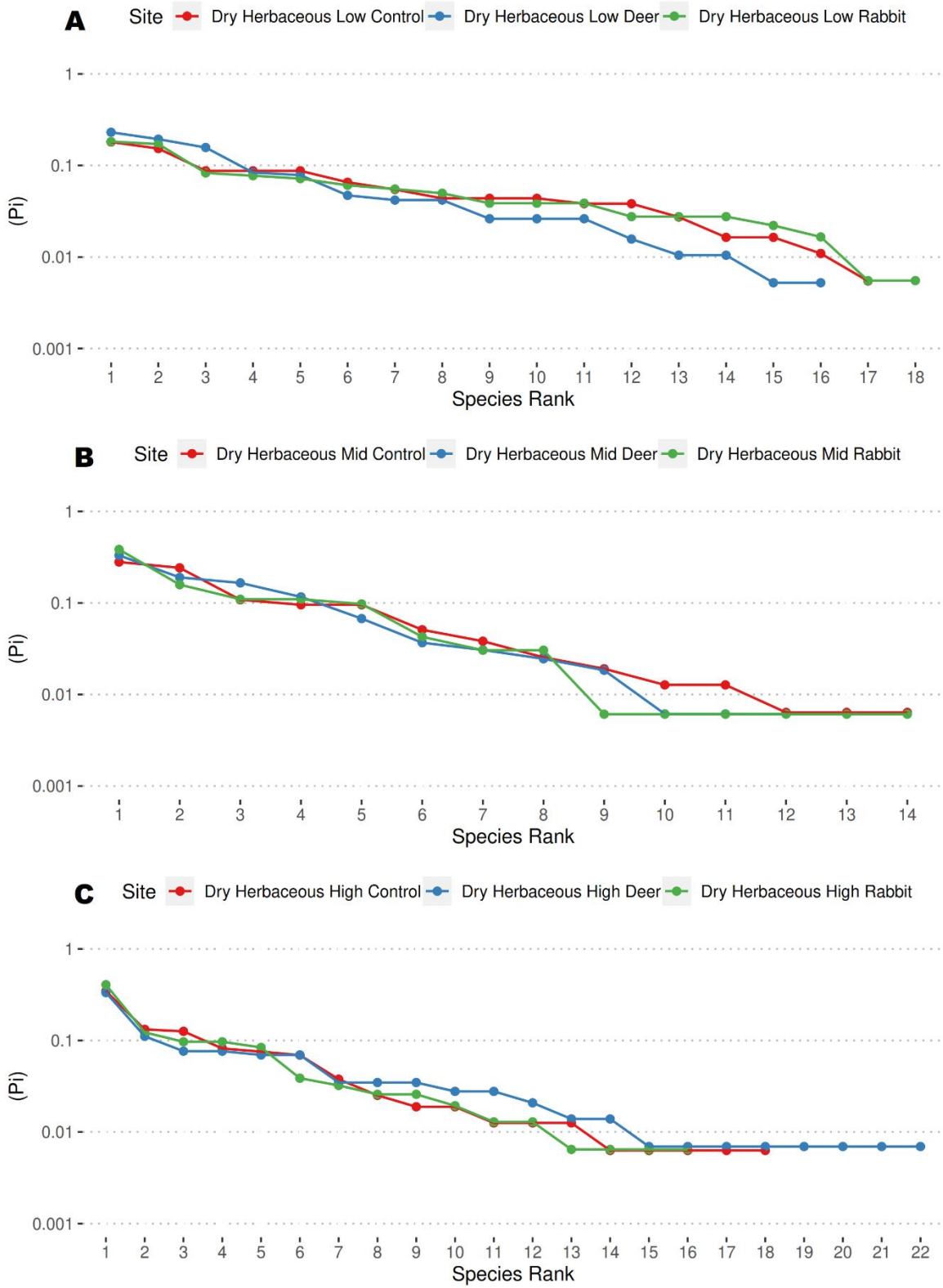


Figure 13. Whittaker curves for all sites in the “Dry Herbaceous Vegetation” habitat

category. (A) “Low” altitudinal category. (B) “Mid” altitudinal category. (C) “High” altitudinal category.

The “Exposed soil” habitat category exhibited fewer plant species, ranging from a minimum of 6 to a maximum of 13 species. The three most dominant plants were *Lupinus microphyllus* (Fabaceae), *Hypochaeris sessiliflora* (Asteraceae), and *Neobartsia pedicularoides* (Scrophulariaceae). Many co-dominant species were observed, including *Poa pauciflora* (Poaceae) and *Nototriche phyllantos* (Malvaceae). The three most rare species were *Ephedra rupestris* (Ephedraceae); *Taraxacum officinale* (Asteraceae), *Plantago sericea* (Plantaginaceae) (Figure 14).

The behavior of the curves was similar across the categories: “Exposed Soil Low” (Figure 14A), “Exposed Soil Mid” (Figure 14B), and “Exposed Soil High” (Figure 14C) all exhibited at least three dominant species, many co-dominant species, and few rare species.

It is important to note that bare ground was present in 25 plots across different habitat categories, but it was most prevalent in the Exposed soil habitat category, occupying up to half of the area in each plot.

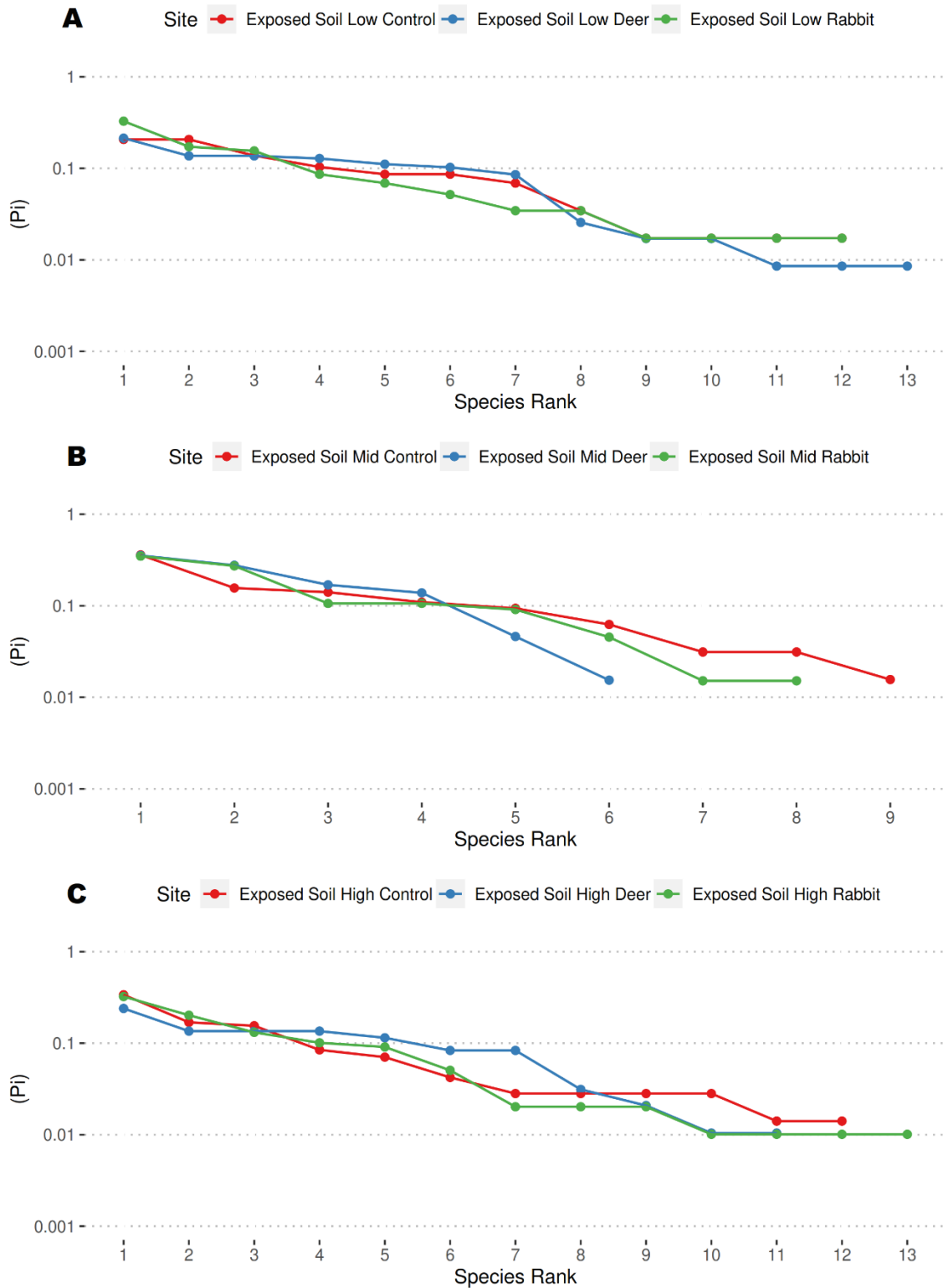


Figure 14. Whittaker curves for all sites in the “Exposed Soil” habitat category. (A) “Low” altitudinal category. (B) “Mid” altitudinal category. (C) “High” altitudinal category.

The results for the "Grassland vegetation" habitat category plots (Figure 15) revealed one to three dominant species, with *Cinnagrostis intermedia* (Poaceae) typically being the most dominant species. In "Grassland Low" (Figure 15A), there were approximately 6 co-dominant and 5 rare species. "Grassland Mid" (Figure 15B) exhibited three dominant species: *Werneria nubigena* (Asteraceae), *Valeriana rigida* (Valerianaceae), and *Agrostis breviculmis* (Poaceae). "Grassland High" (Figure 15C) plots displayed a particularly elevated relative abundance of a single species, namely *Cinnagrostis intermedia* (Poaceae). Meanwhile, co-dominant species were the most numerous in this habitat, with only a few rare species observed, including *Stachys grandidentata* (Lamiaceae) and *Stellaria recurvata* (Caryophyllaceae).

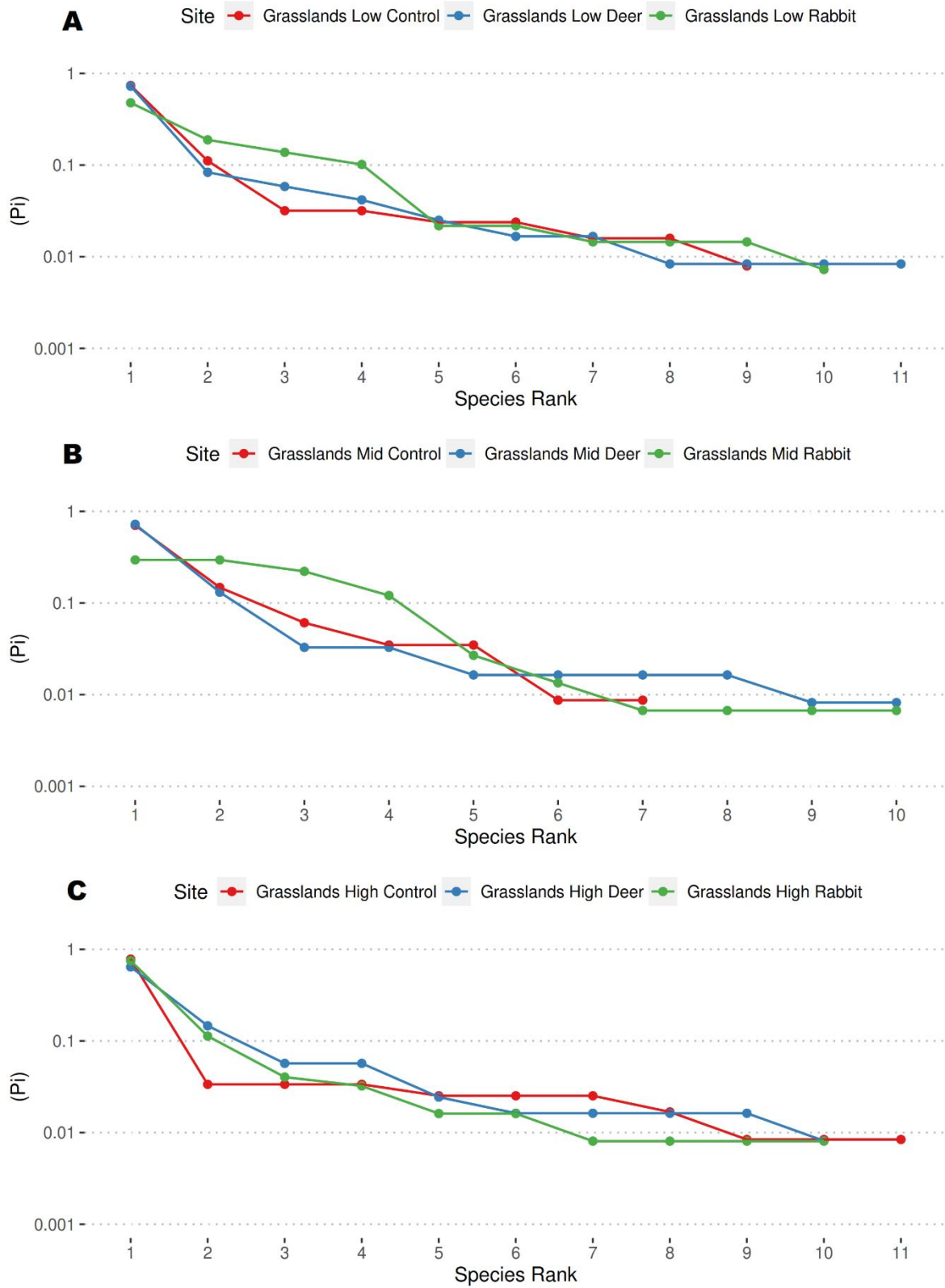


Figure 15. Whittaker curves for all sites in the “Grassland” habitat category. (A) “Low” altitudinal category. (B) “Mid” altitudinal category. (C) “High” altitudinal category.

The results for the "Humid Herbaceous Vegetation" habitat categories were similar. The two most dominant plant species were *Alchemilla propinqua* (Rosaceae), and *Bromus lanatus* (Poaceae). Many co-dominant species were observed, including *Agrostis foliata* (Poaceae), and *Azorella pedunculata* (Apiaceae). The three rare species were *Bidens andicola* (Asteraceae), *Plantago linearis* (Plantaginaceae), and *Gentianella cerastioides* (Gentianaceae) (Figure 16).

In this case, all the plots in the "Humid Herbaceous Low" (Figure 16A), "Humid Herbaceous Mid" (Figure 16B), and "Humid Herbaceous High" (Figure 16C) categories exhibited similar behavior, with a few dominant and rare species but many co-dominant species. The number of rare species varied for each plot.

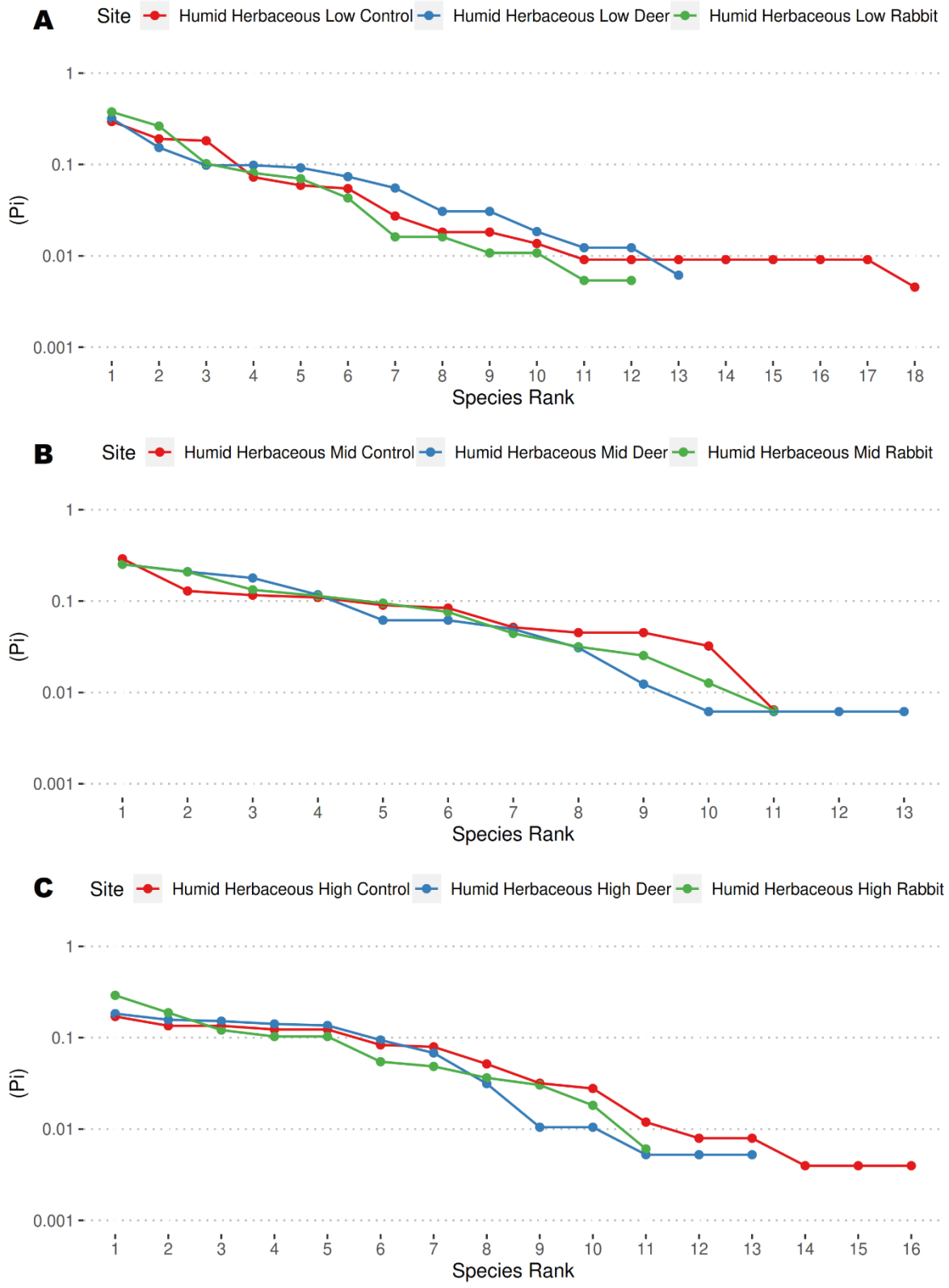


Figure 16. Whittaker curves for all sites in the “Humid Herbaceous Vegetation” habitat category. (A) “Low” altitudinal category. (B) “Mid” altitudinal category. (C) “High” Altitudinal category.

The “Shrubby Vegetation” (Figure 17) habitat category had the richest plot, “Shrubby Vegetation High” (17C). The two most dominant plants were *Azorella pedunculata* (Apiaceae), *Werneria nubigena* (Asteraceae), and *Chuquiraga jussieui* (Asteraceae). There were many co-dominant species in “Shrubby Vegetation Mid” (Figure 17B), for example, *Geranium cerastioides* (Geraniaceae) and *Baccharis caepitosa* (Asteraceae). The three rarest species were *Bidens andicola* (Asteraceae), and *Plantago linearis* (Plantaginaceae) (Figure 17A) in “Shrubby Vegetation Mid.”

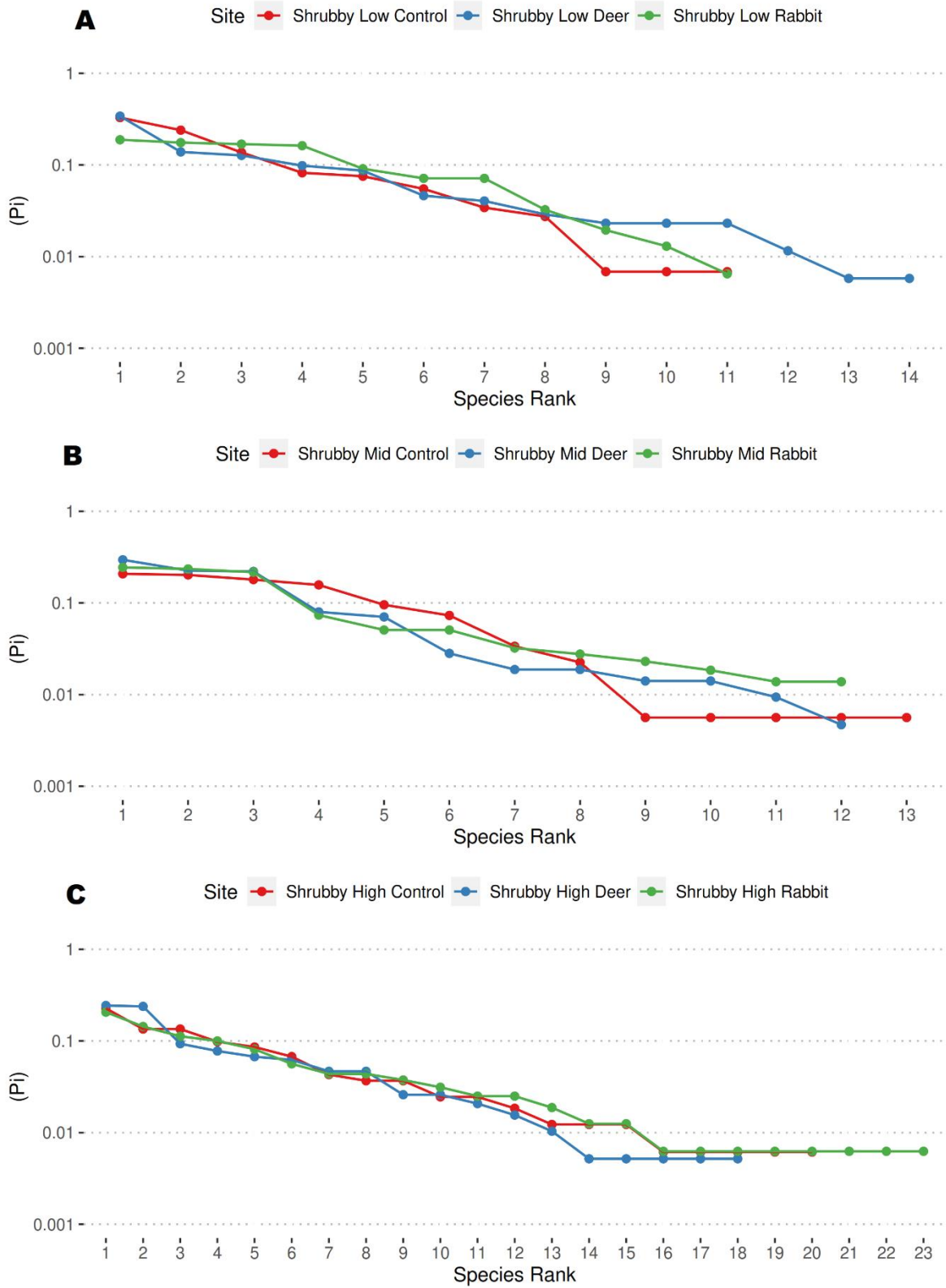


Figure 17. Whittaker Curves for all sites in the “Shrubby Vegetation” habitat category. (A) “Low” altitudinal category. (B) “Mid” altitudinal category. (C) “High” altitudinal category.

4.1.6. Species accumulation curves

The Species accumulation curves were created for each habitat category. Each graph shows the red line with gray confidence intervals, representing how species richness accumulates with increasing sampling effort. The colored boxplots provided a reference distribution of species counts that might be expected by chance (random sampling). This comparison helps assess whether the observed species accumulation in the sampled areas is typical of random sampling.

In the “Dry Herbaceous Vegetation” (Figure 19A), the curve reached a value of 33 species. In the “Humid Herbaceous Vegetation” habitat category (Figure 19B), the curve reached a value of 29 species. The curve in the “Shrubby Vegetation” habitat category (Figure 19C) reached 33 species. In the “Exposed soil” habitat category (Figure 20A), the curve reached a value of 27 accumulated species. The “Cushion plants” category (Figure 20B) presented a curve with 33 accumulated species. Finally, the “Grassland” (Figure 20C) habitat category displayed 19 accumulated species.

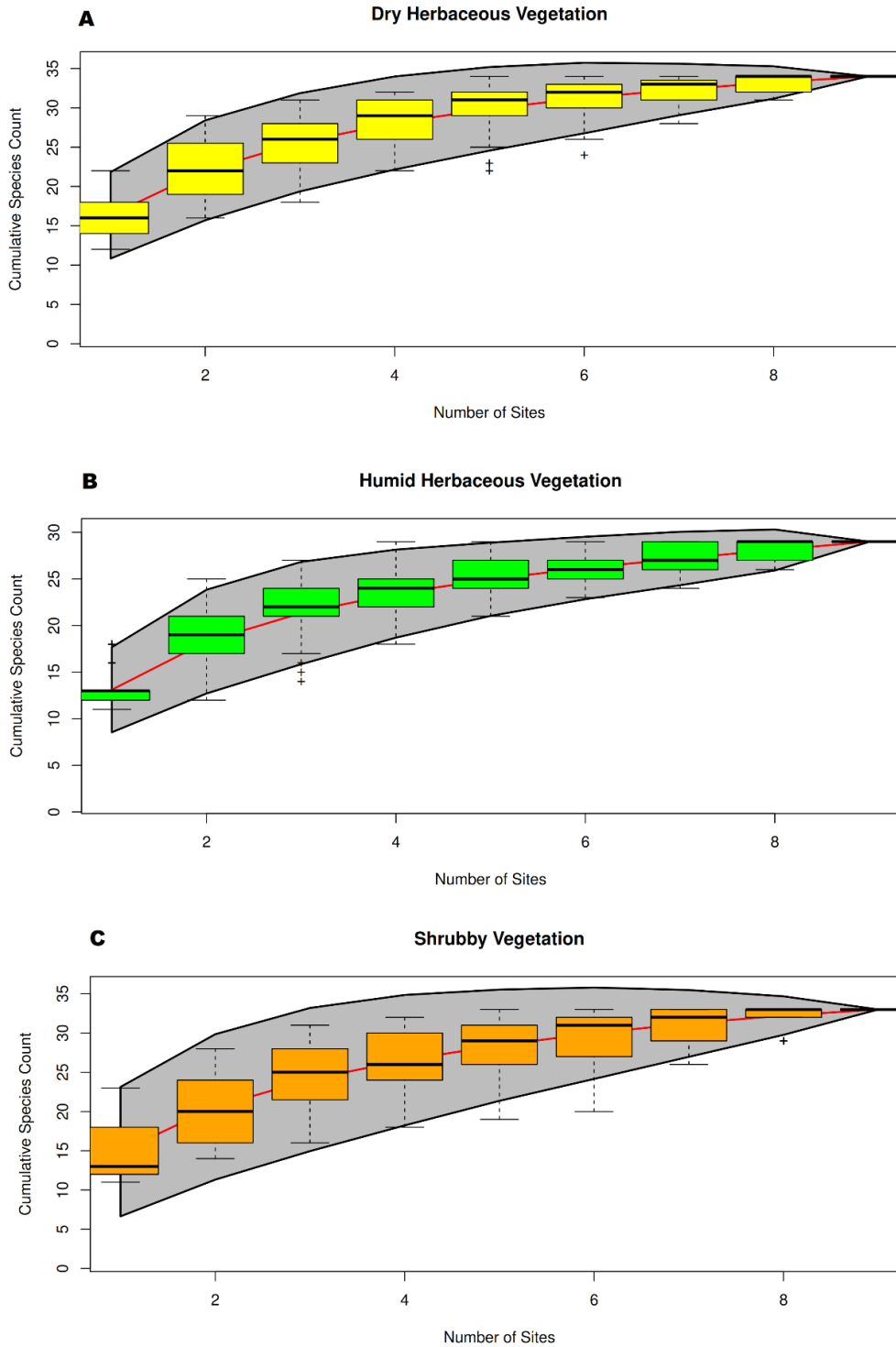


Figure 19. Species Accumulation Curve in Plant Communities from ACHA: Comparing Observed and Randomized Sampling. (A) Species Accumulation Curve for the “Dry Herbaceous Vegetation” Habitat Category. (B) Species Accumulation Curve for the Humid Herbaceous Plants Habitat Category. (C) Species Accumulation Curve for the Shrubby Plants Habitat Category.

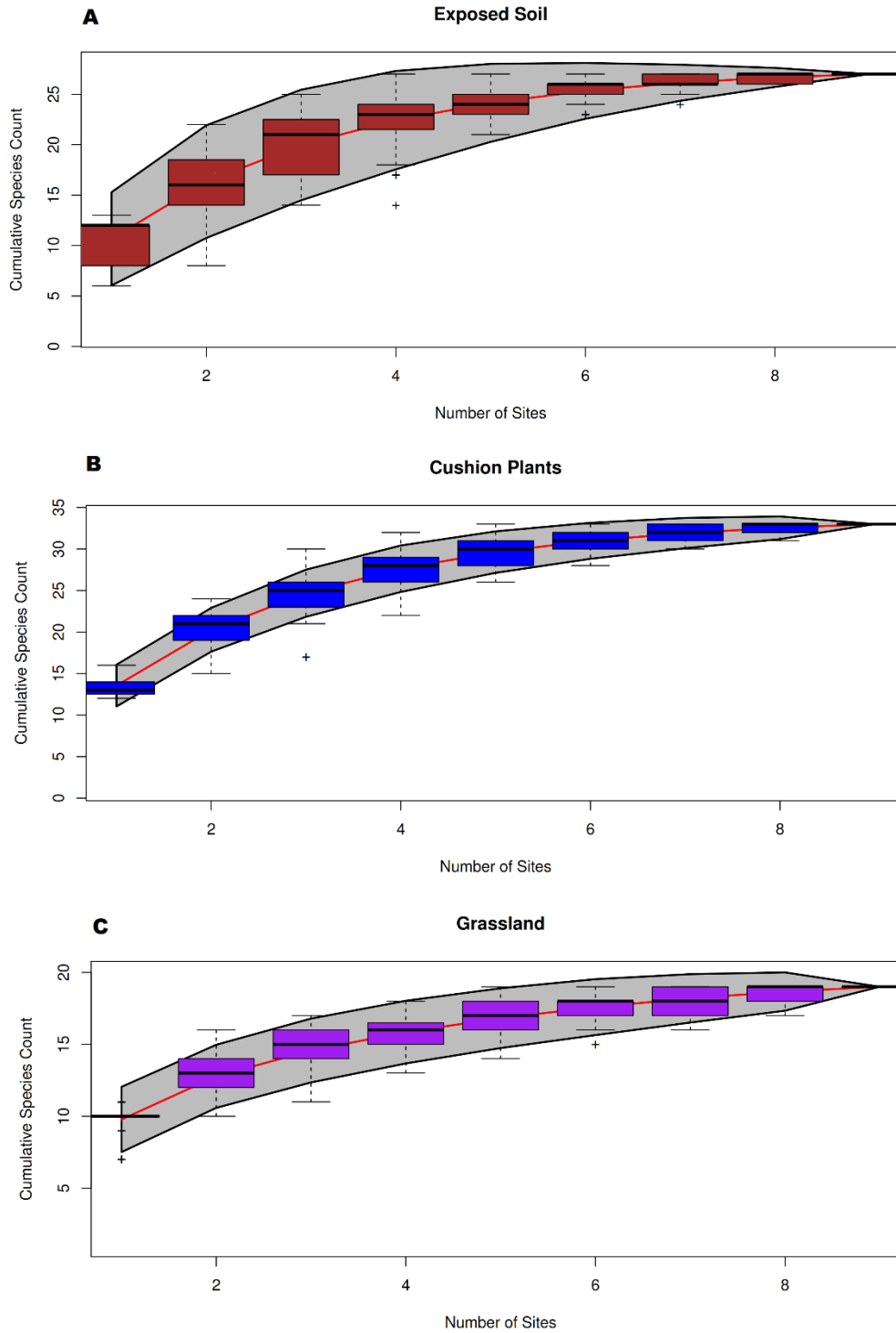


Figure 20. Species Accumulation Curve in Plant Communities: Comparing Observed and Randomized Sampling. (A) Species Accumulation Curve for the Exposed Soil Habitat Category. (B) Species Accumulation Curve for the Cushion Plants Habitat Category. (C) Species Accumulation Curve for the Grasslands habitat category.

4.2. Macro-fungal Characterization

4.2.1. Morphological description

The description of the macrofungal fruiting bodies was performed for all the specimens. The "Exposed Soil" habitat category information was important since many fruiting bodies of *Hygrocybe nigrescens* were found in and around the plots. The observation was at the site called "La Ovejera," where most of the mushrooms were found growing in clusters next to *Hypochaeris sessiliflora* plants (Figure 20B). The main morphological features were a Pileus 8-18 mm diam, parabolic in the margin and umbonate at the apex, convex to conical in mature specimens, color when immature: yellow-red (Figure 20A), color when mature red-brown orange (Figure 20C), surface smooth with a fibrillose margin, margin non-striate. Lamellae are subdisant with color from yellow to white to grey, semi-discolored. The spores were brown with a size of less than 10 μm (Figure 20D). The rest of the macrofungal collection and the corresponding description were stored in the Yachay Botanical Garden.

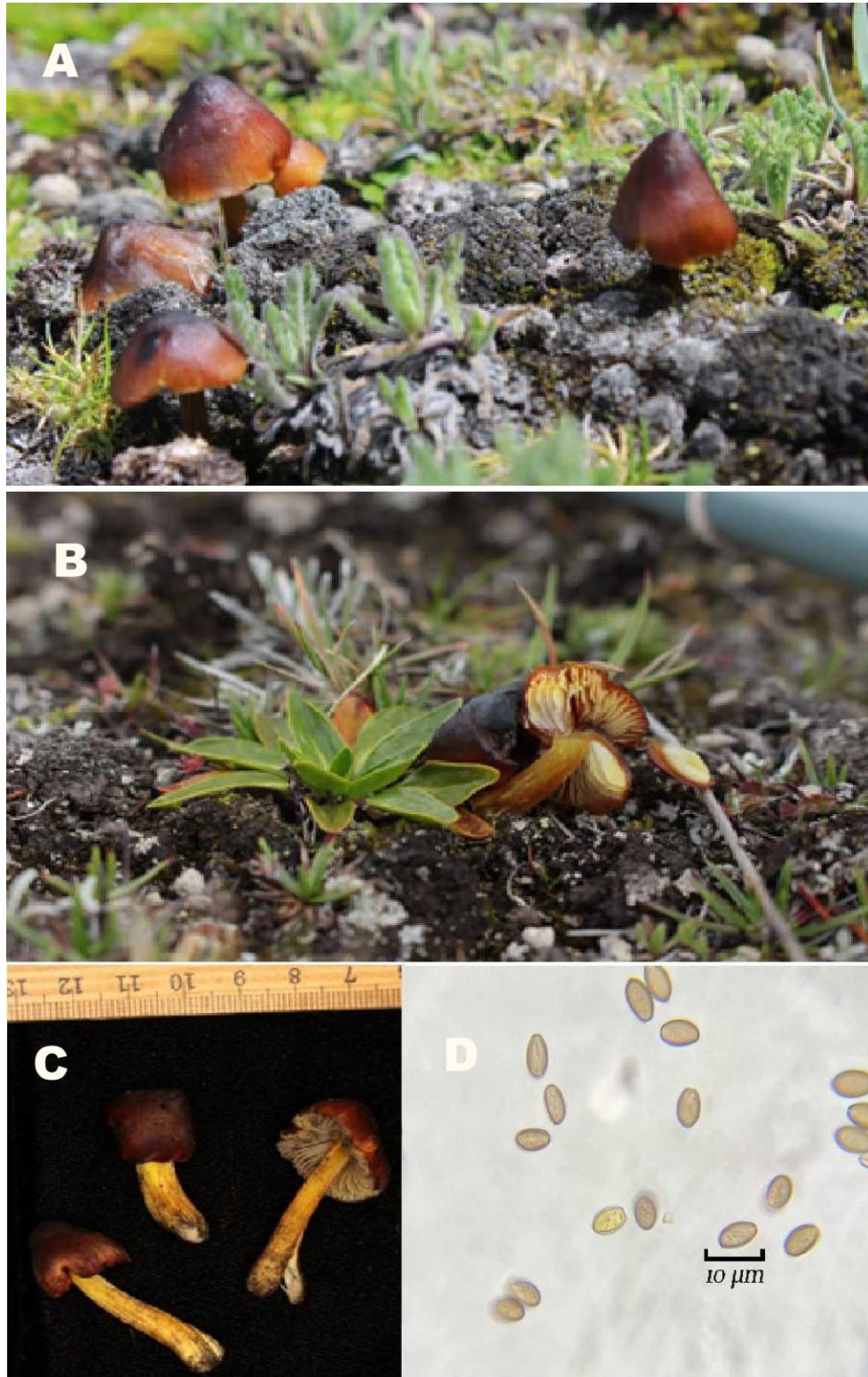


Figure 21. *Hygrocybe nigrescens* in ACHA: (A) Cluster of *Hygrocybe nigrescens*. (B) Mushroom growing from under a *Hypochaeris sessiliflora* plant. (C) Examination under stereoscope (D) Spores under optical microscope with 1000x.

4.2.2. Macro-fungal Richness

From all 54 study plots, six macro-fungal species growing inside the plots were identified (Table 3). They belonged to the Basidiomycota phyla and the order Agaricales.

Table 3. Macro-fungal specimens found in the study plots.

Specie	Family	Order	Pinlum	Site Label	Ground Cover cm ²
<i>Armillaria sp</i>	Physalacriaceae	Agaricales	Basidiomycota	HLC	4.3
<i>Bovista nigrescens</i>	Lycoperdaceae	Agaricales	Basidiomycota	DLR	1.8
<i>Bovista plumbea</i>	Lycoperdaceae	Agaricales	Basidiomycota	SMR	2.4
<i>Conocybe lactea</i>	Bolbitaceae	Agaricales	Basidiomycota	CLR	3
<i>Galerina marginata</i>	Hymenogastraceae	Agaricales	Basidiomycota	GLC	2.4
<i>Hygrocybe nigrescens</i>	Hygrophoraceae	Agaricales	Basidiomycota	ELC	7.7

The Basidiomycete *Armillaria sp.* is a plant pathogen that affects the roots of plants (Figure 22A). This mushroom is very palatable for white-tailed deer (Albuja, 2007), and it was found in the “Humid Herbaceous Low Control” plot (Table 3). Two Gasteroid fungi were identified *Bovista nigrescens* was found in the “Dry Herbaceous Low Rabbit” plot (Figure 22F)., and *Bovista plumbea* was found in the “Shrubby Mid Rabbit” plot (Figure 22E). *Conocybe lactea* was found in the “Cushions Low Rabbit” plot (Figure 22C) and *Galerina marginata* in the “Grassland Low Control” plot (Figure 22D), these mushrooms were toxic to humans but safe for deer consumption. *Hygrocybe nigrescens* was the most abundant mushroom in this report, and it was found in the “Exposed Soil Low Control” (Figure 22B).

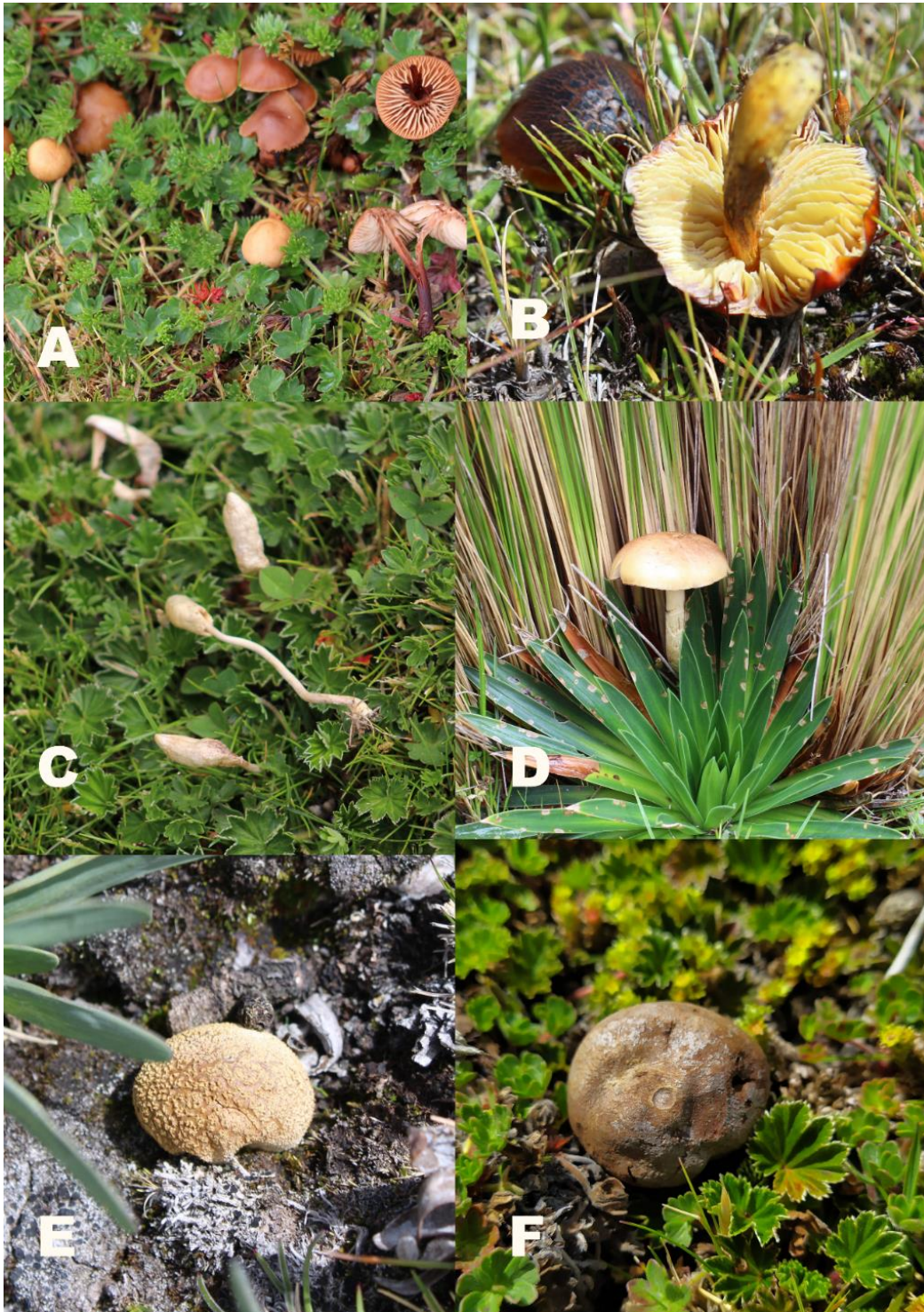


Figure 22. Macrofungal Diversity in All Study Sites in ACHA. (A) *Armillaria* sp. (B) *Hygrocybe nigrescens* (C) *Conocybe lactea* (D) *Galerina marginata* (E) *Bovista plumbea* (F) *Bovista nigrescens* (Photos by: Bryan Israel Vásquez Méndez)

5. Discussion

The Paramo landscapes in the region predominantly comprise large areas of dominant tall-tussock species (e.g., *Calamagrostis intermedia*). However, in ACHA, big patches of Azonal cushion plants (e.g., *Azorella pedunculata* and *Plantago rigida*) and herbaceous vegetation (e.g., *Werneria nubigena* and *Hypochaeris sessiliflora*) were prevalent. These characteristic habitats in ACHA seem to be the product of the intense cattle overgrazing in previous years, coupled with the common practice of burning páramo (Grubb et al., 2020), which acted as a strong selective factor favoring the expansion of certain non-palatable or fire-resistant plant species. (Velásquez-Romo, 2000) A study described the species indicators of overgrazed and fire-disturbed sites (e.g., *Eryngium humile*, *Hypochaeris sessiliflora*, *Bidens andicola*, and *Bromus lanatus*), which were dominant and co-dominant in some of the plots of this study.

Furthermore, various authors (Buytaert et al., 2006; Suárez et al., 2013; Paez-Bimos et al., 2022) have demonstrated the relationship between vegetal cover and hydrological regulation in Antisana. For example, Páez-Bimos et al., (2022) determined the influence of soil-vegetation associations on soil water balance on two soil profiles under different vegetation types. They found that under cushion plants (e.g., *Azorella pedunculata*) with a coarser root system, there was a related increase in soil porosity and higher total available water storage. In contrast, under tall-tussock grassland vegetation (e.g., *Cinnagrostis intermedia*) with a finer and deeper root system, less total available water was observed. Additionally, the significant role of tall tussock grasses and small shrubs in soil protection from water evaporation on bare soil surfaces has been highlighted (Paez-Bimos et al., 2022). Another study (Suárez et al., 2013) reported that water infiltration was extremely high under shrubby vegetation and decreased markedly under grassland vegetation.

Studies conducted to analyze the impact of burning, grazing, and trampling (Grubb et al., 2020) emphasize the long-term effects of herbivores on vegetative composition. Evidence showed that with the removal of livestock in the Antisana National Park, the decrease in grazing pressure resulting from the absence of cattle and sheep has had a positive effect on vegetative coverage. However, the impacted areas are still recovering (Ramirez et al., 2019). Today, the most significant herbivory pressure is posed by white-tailed deer. Deer can significantly influence vegetation structure and composition, particularly when deer populations become overabundant. A recent study on the density of white-tailed deer in ACHA (Figure 3) has documented increasing numbers in recent years (Tellkamp et al., 2019). This study underscores the necessity of analyzing the effects of rising herbivore numbers on vegetal composition at the site level in ACHA.

The initial estimation of deer abundance, reported by Tellkamp et al. (2019), indicates a density of 11.27 individuals per square kilometer for ACHA, which is significantly higher than the density reported for an area near ACHA called Guaytaloma, with 1.6 individuals per square kilometer, as reported by Albuja (2007).

Despite the density reported in ACHA not being as high compared to other sites in Colombia or Venezuela, the number of deer that the reserve should hold depends on the management goals of this area. This management is contingent upon the combination of vegetation that favors the greatest levels of water retention and the slowest rates of water release, as well as how deer contribute to affecting them in the long term (Tellkamp et al., 2019).

Additionally, authors have demonstrated a correlation between vegetation cover and the densities of small herbivores, such as the Andean rabbit (*Sylvilagus andinus*) (Figure 22) (J. García et al., 2016), attributed to herbivory, scratching, digging, and nutrient distribution by feces. However, the density of Andean rabbits in ACHA has fluctuated, possibly due to factors such as viral diseases like the Myxoma virus and rabbit hemorrhagic disease, as reported by Arias et al. (2021). Another contributing factor could be the increasing number of predators. According to park rangers, sightings of large predators have been higher in recent years,

including birds such as the great horned owl (*Bubo virginianus*) (Figure 1B), the Andean fox (*Lycalopex culpaeus*) (Figure 1E), and puma (*Puma concolor*) (M. Simba, personal communication, July 2023).



Figure 23. Andean rabbit (*Sylvilagus andinus*) in ACHA. (Photo by: Bryan Israel Vásquez Méndez).

The next step in this 4-year-long project includes an Exclusion experiment (Annex 2), which has been conducted in different ecosystems and for various purposes. For instance, White-tailed deer enclosure studies in Pennsylvania were carried out in an oak forest to evaluate timber production. In Japan, a deer exclusion study in an old-growth forest reported the response of forest floor vegetation and tree regeneration (Nomiya et al., 2002). Similarly, in Madrid, Spain, the impact of deer on Mediterranean shrublands was assessed, focusing on woody plant diversity (Perea et al., 2014).

These experiments (McCormick et al., 1993; Hofstede, 1995; Nomiya et al., 2002; Côté et al., 2004; DiTommaso et al., 2014; Ramirez et al., 2019) vary in the number and size of exclusions, as well as the duration, which ranges from 3 to 7 years. However, they all coincide in revealing the negative effect of deer overabundance on vegetal composition after the exclusion treatments attributed to intense herbivory (Figure 23B) and trampling (Figures 23A and 23C). In other ecosystems, deer overabundance threatens vegetation dynamics by forcing the vegetation succession to earlier stages. However, there is currently no clear data on how White-tailed deer affect vegetation structure and composition in ACHA. Therefore, it is essential to closely monitor the number of deer and the vegetal composition in order to understand their relationship.



Figure 24. Herbivory in ACHA. (A) and (C) Evidence of trampling, (B) Evidence of grazing. (Photo by: Bryan Israel Vásquez Méndez).

Dietary studies conducted on white-tailed deer in North America have confirmed the significant contribution of mushrooms to cervids as a source of nutrients, including phosphorus, potassium, selenium, and iron, as well as vitamins such as thiamine and riboflavin (Launchbaugh, 1992; Cadotte et al., 2021). Conversely, by consuming mushrooms, the feces of the cervid serve as a vector for the dispersal of mushroom spores (Cadotte et al., 2021). Furthermore, spores and mycelium of some species contain a molecule called glomalin, which acts as a natural glue for soil particles, preventing soil erosion (Singh et al., 2020).

Interactions between vascular flora and mycorrhizal fungi play a pivotal role in determining the formation and diversity of plant communities (Moora, 2014). Many authors have highlighted the common occurrence of mycorrhization in native Andean plants, which enhances nutrient uptake through mycelium networks (Casanova et al., 2011). Several studies have documented the transfer of minerals such as nitrogen (Siddiqui et al., 2008) and phosphorus (Brundrett, 2008), as well as water transport (Marjanaovic & Nehls, 2008), contributing to enhanced plant resistance to drought during the dry season (Molina-Montenegro et al., 2015). Another study describes habitats dominated by cushion plants had an incremented number of microbial communities like arbuscular mycorrhizal fungi (Rodríguez-Echeverría et al., 2021).

The current literature suggests that neighboring plants share mycorrhizal networks, which can have both positive and negative effects on plant communities (Molina-Montenegro, 2015). Negative interactions, such as those caused by pathogenic fungi leading to root system rot (Casanova et al., 2011), can also alter plant composition. Additionally, saprophytic mushrooms degrade organic matter and increase nutrient availability in the soil.

Despite the importance of macrofungal organisms in páramo ecosystems, there is limited documentation, with only a single record (e.g., *Bovista plumbea*) for the Antisana Hydric Conservation Area (ACHA) in the fungi collection of the National Herbarium of Ecuador (QCNE) (Molina et al., 2020). Recognizing macrofungal specimens as a fundamental component of the ecosystem is crucial

due to their role in providing dietary support for white-tailed deer and serving as the underground network that connects plant communities (García et al., 2004). Currently, it is well-recognized that plant communities tend to share mycorrhizal networks. Furthermore, fungal associations can improve plant survival and establishment in harsh environments (Molina-Montenegro et al., 2015). A more interdisciplinary approach integrating fungi into the discussion will lead to a better understanding of this fragile ecosystem and possibly improve restoration techniques.

Plant identification was a crucial part of this initial characterization, which is not optimal with only a plant checklist. For this reason, a páramo plants field guide is being completed to aid in plant identification in future characterizations for the herbivory project and other botanical endeavors inside ACHA (Figure 25).

Glaciers in the tropics are very sensitive to climate change. Runoff from glaciers increases as temperature goes up, and downstream reservoirs can overflow, causing floods during the rainy season. (Vuille et al., 2008). The accelerating increases in species richness on mountain summits across the world demonstrate that biotic change is occurring in the most remote places on Earth, with potentially long-lasting consequences for biodiversity and ecosystem services, as explained by Steinbauer et al. (2018), these effects could get accentuated by global warming.

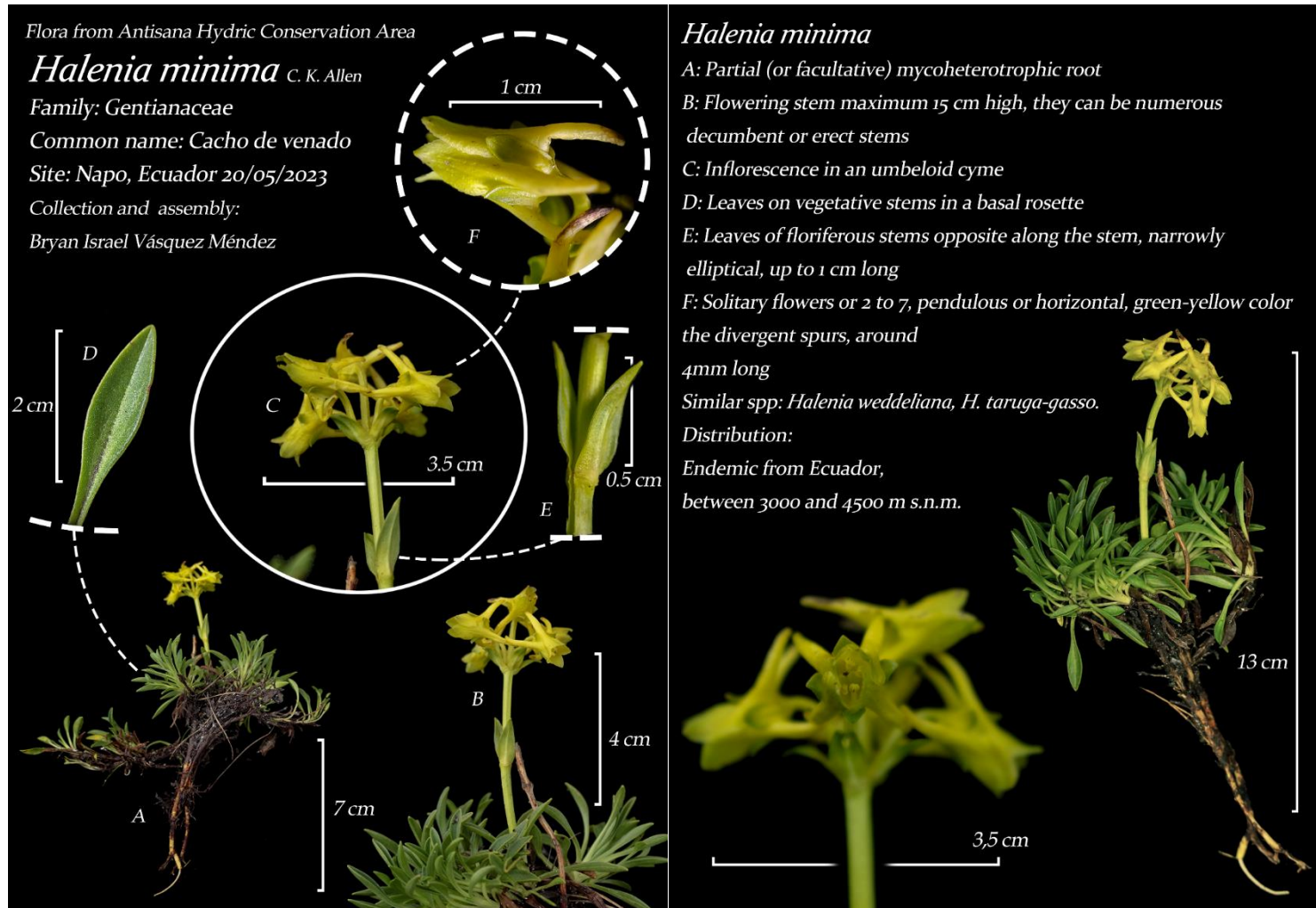


Figure 25. Lankester Composite Dissection Plates applied to páramo vegetation. *Halenia minima* (Gentianaceae) “Vulnerable” conservation status (Photos by: Bryan Israel Vásquez Méndez).

5. Conclusions

In conclusion, this study reports the characterization of the flora dominance and diversity in ACHA, alongside the presence of Macro-fungal organisms was described. I established N=54 plots (3m x 2m) to sample tracheophytes in this area. Exclusions were constructed in some of the plots, and their effect will be evaluated over time and compared with this initial report. The following habitats were considered: Shrubby, Exposed Soil, Cushions, Grasslands, Dry herbaceous, and Humid Herbaceous. Dominance, measured as the relative vegetation cover percentage, was estimated using the square-point method for each species. A total of 69 species of tracheophytes were identified, represented by 47 genera and 23 families. Among the 69 species, twelve were endemic, among which five were classified with a conservation status of "Least Concern" (LC), two (*Distichia acicularis*, Juncaceae; *Drava obovata*, Brassicaceae) were considered "Near Threatened" (NT), and three (*Castilleja nubigena*, Scrophulariaceae; *Carex toreadora*, Cyperaceae; *Geranium sericeum*, Geraniaceae) were considered "Vulnerable" (VU). The 56 left are native, and one (*Taraxacum officinale*, Asteraceae) is classified as introduced.

The three most species-rich plots were: "Shrubby High Rabbit" with 23 species, "Dry Herbaceous High Deer" with 22 species, and "Shrubby High Control" with 20 species. The three least species-rich were: "Exposed Soil Mid Deer" with 6, "Grasslands Mid Control" with 7, and "Exposed Soil Mid Rabbit" with 8 species.

The nonlinearity of diversity indices can lead to errors if compared directly. So, to compare the diversity of different plots, the Effective Number of Species based on the Shannon Diversity Index was obtained. The three most diverse plots, according to the Effective Number of Species based on the Shannon Diversity Index, were "Shrubby High Rabbit" (13 effective spp.), "Dry Herbaceous Low Rabbit" (13 effective spp.), and "Dry Herbaceous Low Control" (13 effective spp). Conversely, the least diverse plots according to the same Index were "Grasslands High Rabbit"

(3 effective spp.), "Grasslands High Control" (3 effective spp.), and "Grasslands Mid Control" (3 effective spp.).

The Rank abundance curves for the three plots in each Habitat and Altitudinal category were displayed in a single graph to analyze the abundance of plant species. It is important to note that bare ground was present in 25 plots across different habitat categories, but it was most prevalent in the Exposed soil habitat category.

The Distance-based Multivariate Analysis revealed that the abundance data was clustered differently for each habitat category. Specifically, the "Exposed Soil" habitat category was separated from the rest of the clusters, while the "Grassland" and "Cushion vegetation" habitat categories were clustered at opposite ends of the ordination. Conversely, the "Dry Herbaceous vegetation," "Humid Herbaceous vegetation," and "Shrubby vegetation" habitat categories overlapped in the middle of the dissimilarity matrix.

From all 54 study plots, six macro-fungal species growing inside the plots were identified. They belonged to the Basidiomycota phyla and the order Agaricales. The Basidiomycete *Armillaria* sp. is a plant pathogen that affects the roots of plants. This mushroom was very palatable for white-tailed deer, and it was found in the "Humid Herbaceous Low Control" plot. Two Gasteroid fungi were identified *Bovista nigrescens* was found in the "Dry Herbaceous Low Rabbit" plot., and *Bovista plumbea* was found in the "Shrubby Mid Rabbit" plot. *Conocybe lactea* was found in the "Cushions Low Rabbit" plot and *Galerina marginata* in the "Grassland Low Control" plot. These mushrooms were toxic to humans but safe for deer consumption. *Hygrocybe nigrescens* was the most abundant mushroom in this report, and it was found in the "Exposed Soil Low Control" Plot.

This is the first report at the site level to explore the floristic dominance and diversity in habitats visited by the white-tailed deer. These results are a solid base to compare future data. This is my contribution to conserving this majestic place.

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6. Annex

Annex 1. The table with the names, labels, and coordinates of the plots.

Name of the plot	Label	UTM Coordinates
Humid Herbaceous Low Deer Exclusion Plot	HLD	17 M 807118 9941558
Humid Herbaceous Low Rabbit Exclusion Plot	HLR	17 M 807192 9941506
Humid Herbaceous Low Control Plot	HLC	17 M 807159 9941539
Humid Herbaceous Mid Deer Exclusion Plot	HMD	17 M 806938 9942033
Humid Herbaceous Mid Rabbit Exclusion Plot	HMR	17 M 806919 9942092
Humid Herbaceous Mid Control Plot	HMC	17 M 806885 9942095
Humid Herbaceous High Deer Exclusion Plot	HHD	17 M 806216 9942438
Humid Herbaceous High Rabbit Exclusion Plot	HHR	17 M 806258 9942492
Humid Herbaceous High Control Plot	HHC	17 M 806224 9942487
Dry Herbaceous Low Deer Exclusion Plot	DLD	17 M 807600 9943478
Dry Herbaceous Low Rabbit Exclusion Plot	DLR	17 M 807590 9943446
Dry Herbaceous Low Control Plot	DLC	17 M 807546 9943476
Dry Herbaceous Mid Deer Exclusion Plot	DMD	17 M 809776 9944868
Dry Herbaceous Mid Rabbit Exclusion Plot	DMR	17 M 809789 9944819
Dry Herbaceous Mid Control Plot	DMC	17 M 809800 9944847
Dry Herbaceous High Deer Exclusion Plot	DHD	17 M 812729 9946991
Dry Herbaceous High Rabbit Exclusion Plot	DHR	17 M 812700 9946918
Dry Herbaceous High Control Plot	DHC	17 M 812699 9946844
Cushion Low Deer Exclusion Plot	CLD	17 M 805647 9942486

Cushion Low Rabbit Exclusion Plot	CLR	17 M 805662 9942414
Cushion Low Control Plot	CLC	17 M 805680 9942372
Cushion Mid Deer Exclusion Plot	CMD	17 M 810079 9944451
Cushion Mid Rabbit Exclusion Plot	CMR	17 M 810111 9944494
Cushion Mid Control Plot	CMC	17 M 810136 9944467
Cushion High Deer Exclusion Plot	CHD	17 M 812399 9947613
Cushion High Rabbit Exclusion Plot	CHR	17 M 812369 9947737
Cushion High Control Plot	CHC	17 M 812505 9947546
Grassland Low Deer Exclusion Plot	GLD	17 M 810088 9942969
Grassland Low Rabbit Exclusion Plot	GLR	17 M 810048 9942972
Grassland Low Control Plot	GLC	17 M 810067 9942923
Grassland Mid Deer Exclusion Plot	GMD	17 M 805760 9946290
Grassland Mid Rabbit Exclusion Plot	GMR	17 M 805727 9946295
Grassland Mid Control Plot	GMC	17 M 805725 9946345
Grassland High Deer Exclusion Plot	GHD	17 M 809745 9947756
Grassland High Rabbit Exclusion Plot	GHR	17 M 809757 9947711
Grassland High Control Plot	GHC	17 M 809757 9947712
Shrubby Low Deer Plot	SLD	17 M 809519 9947218
Shrubby Low Rabbit Plot	SLR	17 M 809511 9947163
Shrubby Low Control Plot	SLC	17 M 809447 9947154
Shrubby Mid Deer Exclusion Plot	SMD	17 M 805761 9947121
Shrubby Mid Rabbit Exclusion Plot	SMR	17 M 805814 9947113
Shrubby Mid Control Plot	SMC	17 M 805791 9947243

Shrubby High Deer Exclusion Plot	SHD	17 M 812438 9946579
Shrubby High Rabbit Exclusion Plot	SHR	17 M 812485 9946612
Shrubby High Control Plot	SHC	17 M 812388 9946522
Exposed Soil Low Deer Exclusion Plot	ELD	17 M 808599 9944271
Exposed Soil Low Rabbit Exclusion Plot	ELR	17 M 808609 9944337
Exposed Soil Low Control Plot	ELC	17 M 808554 9944304
Exposed Soil Mid Deer Exclusion Plot	EMD	17 M 808304 9946756
Exposed Soil Mid Rabbit Exclusion Plot	EMR	17 M 808304 9946786
Exposed Soil Mid Control Plot	ESMC	17 M 808267 9946808
Exposed Soil High Deer Exclusion Plot	ESHD	17 M 812159 9947388
Exposed Soil High Rabbit Exclusion Plot	EHR	17 M 812156 9947341
Exposed Soil High Control Plot	EHC	17 M 812136 9947418

Annex 2. Exclusions designs and placement in ACHA.

Fence location to exclude deer and rabbits from study plots (Exclusion plots) is a proven way to bring an understanding of herbivore-vegetation interactions (Côté et al., 2004; Gómez, 2017). Exclusion plots were placed systematically in randomly selected places. Parameters for the placement of every plot included the altitudinal gradient, presence of characteristic species, avoidance of big slopes to prevent seed rain and shadow, and at least 50 meters away from roads, rivers, and habitat margins. The fences were installed with the assistance of 10 guardapáramos from FONAG and 20 students from Yachay Tech.

Following the recommendations of Vercauteren et al. (2006), the dimensions for all exclusion plots were 1.8 m high, 3 meters long, and 2 meters wide. Each plot has an area of 6 m². The exclusion plots considered two designs: one to exclude White-tailed deer (Figure 5A) and the other to exclude Andean rabbits (Figure 5B). To prevent deer entrance, barbed wire was placed every 20 cm along the posts, and to prevent deer and rabbit entrance, a metallic grid was buried 50 cm underground and extended 50 cm above the ground. A control plot accompanies both exclusions, consisting of red PVC tubes in the vertices.

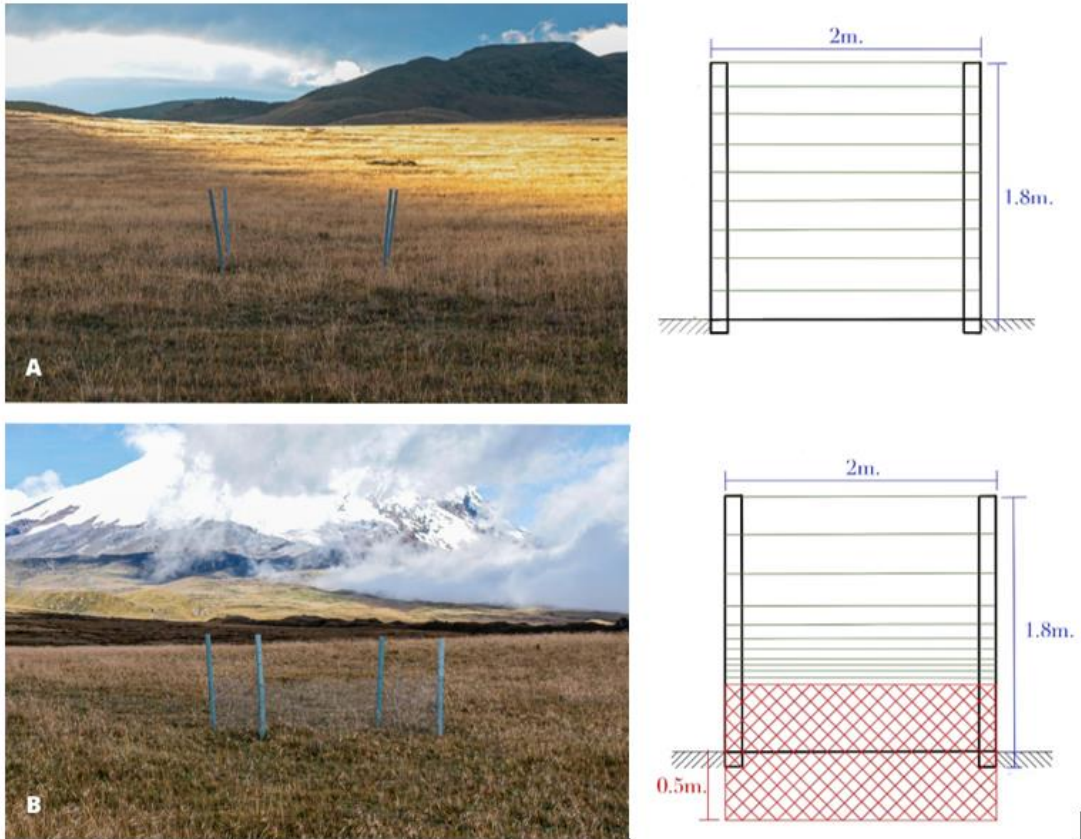


Figure 26: Type of exclusions placed in ACHA with a side view diagram. (A) Deer exclusion. (B) Rabbit exclusion.

Annex 3. Data about species, authors, endemism, and IUCN status for the taxa recorded in the present study.

Species	Author	Status	Status IUCN
<i>Aphanactis ollgaardii</i>	H.Rob.	Endemic	Least concern
<i>Astragalus geminiflorus</i>	Humb. & Bonpl.	Endemic	Least concern
<i>Carex toreadora</i>	Steyerm.	Endemic	Least concern
<i>Castilleja nubigena</i>	Kunth	Endemic	Least concern
<i>Distichia acicularis</i>	Balslev & Laegaard	Endemic	Least concern
<i>Draba obovata</i>	Benth.	Endemic	Near Threatened
<i>Festuca chimborazensis</i>	E.B.Alexeev	Endemic	Near Threatened
<i>Gentianella rupicola</i>	(Kunth) Holub	Endemic	Vulnerable
<i>Geranium sericeum</i>	Willd. ex Spreng.	Endemic	Vulnerable
<i>Halenia minima</i>	C.K.Allen	Endemic	Vulnerable
<i>Stachys grandidentata</i>	Lindl.	Endemic	
<i>Stellaria recurvata</i>	Willd. ex D.F.K.Schltld.	Endemic	
<i>Taraxacum officinale</i>	F.H.Wigg.	<u>Introduced</u>	
<i>Agrostis breviculmis</i>	Hitchc.		Native
<i>Agrostis foliata</i>	Hook.f.		Native
<i>Alchemilla orbiculata</i>	H.Lindb. ex Juz.		Native
<i>Alchemilla uniflora</i>	(Maguire) Govaerts		Native
<i>Azorella aretioides</i>	Willd. ex DC.		Native
<i>Azorella biloba</i>	Wedd.		Native
<i>Azorella pedunculata</i>	(Spreng.) Mathias & Constance		Native
<i>Baccharis caespitosa</i>	Pers.		Native
<i>Bidens andicola</i>	Kunth		Native
<i>Bromus lanatus</i>	Kunth		Native
<i>Calandrinia acaulis</i>	Kunth		Native

Species	Author	Status	Status IUCN
<i>Caltha sagittata</i>	Bercht. & J.Presl	Native	
<i>Carex pichinchensis</i>	Kunth	Native	
<i>Carex pygmaea</i>	Boeckeler	Native	
<i>Castilleja pumila</i>	(Benth.) Wedd.	Native	
<i>Cerastium danguyi</i>	J.F.Macbr.	Native	
<i>Cerastium floccosum</i>	Benth.	Native	
<i>Cerastium imbricatum</i>	Kunth	Native	
<i>Chaerophyllum andicola</i>	(Kunth) K.F.Chung	Native	
<i>Chuiraga jussieui</i>	J.F.Gmel.	Native	
<i>Cinnagrostis coarctata</i>	(Kunth) P.M.Peterson, Soreng, Romasch. & Barberá	Native	
<i>Cinnagrostis intermedia</i>	(J.Presl) P.M.Peterson, Soreng, Romasch. & Barberá	Native	
<i>Cinnagrostis jamesonii</i>	(Steud.) P.M.Peterson, Soreng, Romasch. & Barberá	Native	
<i>Distichia muscoides</i>	Nees & Meyen	Native	
<i>Ephedra rupestris</i>	Benth.	Native	
<i>Erigeron cardaminifolius</i>	Wedd.	Native	
<i>Erigeron ecuadoriensis</i>	Hieron.	Native	
<i>Eryngium humile</i>	Cav.	Native	
<i>Gentiana sedifolia</i>	Kunth	Native	
<i>Gentianella cerastioides</i>	(Kunth) Fabris	Native	
<i>Geranium multipartitum</i>	Benth.	Native	
<i>Geranium reptans</i>	R.Knuth	Native	
<i>Geranium siboldioides</i>	Benth.	Native	
<i>Huperzia crassa</i>	(Humb. & Bonpl. ex Willd.) Rothm.	Native	

<i>Hypochaeris sessiliflora</i>	Kunth	Native
<i>Lasiocephalus ovatus</i>	Schlttdl.	Native
<i>Lucilia kunthiana</i>	(DC.) Zardini	Native
<i>Lupinus microphyllus</i>	Desr.	Native
<i>Lysipomia montioides</i>	Kunth	Native
<i>Neobartsia pedicularoides</i>	(Benth.) Uribe-Convers & Tank	Native
<i>Neobartsia stricta</i>	(Benth.) Uribe-Convers & Tank	Native
<i>Nototriche phyllanthos</i>	A.W.Hill	Native
<i>Ophioglossum crotalophoroides</i>	Walter	Native
<i>Oritrophium crocifolium</i>	(Kunth) Cuatrec.	Native
<i>Oritrophium limnophilum</i>	(Sch.Bip.) Cuatrec.	Native
<i>Plantago linearis</i>	Kunth	Native
<i>Plantago rigida</i>	Kunth	Native
<i>Plantago sericea</i>	Ruiz & Pav.	Native
<i>Poa pauciflora</i>	Roem. & Schult.	Native
<i>Ranunculus praemorsus</i>	Kunth ex DC.	Native
<i>Senecio chionogeton</i>	Wedd.	Native
<i>Valeriana rigida</i>	Ruiz & Pav.	Native
<i>Viola bangii</i>	Rusby	Native
<i>Werneria nubigena</i>	Kunth	Native
<i>Werneria pygmaea</i>	Gillies ex Hook. & Arn.	Native
<i>Xenophyllum humile</i>	(Kunth) V.A.Funk	Native

Annex 5. Small portion of the Codes in RStudio for this Characterization

```
##### CODE FOR PLANT CHARACTERIZATION
#####

# THIS CODE IS OPTIMIZED TO WORK WITH THE DATA BASE OF THE
CHARACERIZATION

# OF THE VEGETAL COMUNITY IN ACHA USING THE SQUARE POINT
METHDOD.

#LOAD THE NECESSARY LIBRARIES

##### LOAD DATA #####

###Set the working directory
setwd("C:/Users/bryan/OneDrive/Esritorio/CODE/CODE_ENG")

###Upload information from the spreadsheet of the square point method
SQUARE_POINT0B <- read_excel("SQUARE_POINT0B.xlsx")
#View(SQUARE_POINT) #See data base
class(SQUARE_POINT0B$'Ground.level') #type of the variable

##### DATA MANAGEMENT #####

VAS<- SQUARE_POINT0B %>%
  select(Site, 'Ground.level', 'Vegetation.level.one','Vegetation.level.two') %>%
  pivot_longer(cols=c('Ground.level', 'Vegetation.level.one','Vegetation.level.two'),
names_to = "level" , values_to= "Species")%>%
  select(Site, Species)

tail(VAS)

### Check the number of species in a site
test<-VAS %>% filter(Site== "Grasslands High Deer") %>%
  filter(!is.na(Species)) %>% distinct(Species)
nrow(test)

## COVER PERCENTAGE FORMULA IS APPLIED HERE:
VAS.sp.abund<-VAS%>%filter(!(is.na(Species))) %>%
  group_by(Site, Species) %>%
  summarise(abundance=n()*(100/96))
```

```

# Abundance is calculated as the count of rows in each group multiplied by (100 /
96)# This calculatio is applied to each group within the dataset
species_total_abundance <- VAS.sp.abund %>%
  group_by(Species) %>%
  summarise(total_abundance = sum(abundance))
print(species_total_abundance)
### Total abundance (N)
total_abundance <- VAS.sp.abund %>%
  group_by(Site) %>%
  summarise(total_abundance = sum(abundance))
summary(total_abundance)
# write_xlsx(total_abundance, "site_total_abundance.xlsx")
# Merge the total abundance data in the Vas.sp.pi dataframe
VAS.sp.pi <- left_join(VAS.sp.abund, total_abundance, by = "Site")
###Pi=(Ns/N)
VAS.sp.pi <- VAS.sp.pi %>%
  mutate(relative_abundance = abundance / total_abundance) %>%
  select(Site, Species, relative_abundance) %>% # Select specific columns from
the original dataframe
  arrange(Site, desc(relative_abundance)) #Order the relative abundance data

```

```

species_total_pi <- VAS.sp.pi %>%
  group_by(Species) %>%
  summarise(total_relative_abundance = sum(relative_abundance))
print(species_total_pi)
### Check ALL PLANT SPECIES PRESENT IN THE DATA BASE
espu<-unique(VAS.sp.abund$Species) # Select only one of the species
print(espu) #and presents them
#####RANK ABUNDANCE PLOT#####
####EXAMPLE WITH CUSHIONS
Almohadillas <- c("Cushions Low Control",
                 "Cushions Low Rabbit",
                 "Cushions Low Deer")
# Filter data for the selected sites
Almohadillas.df <- VAS.sp.pi[grepl(paste0("^", paste(Almohadillas, collapse = "|")),
VAS.sp.pi$Site), ]
# Arrange data by site and relative abundance
Almohadillas.df <- Almohadillas.df %>%
  arrange(Site, desc(relative_abundance))
# Calculate a unique x-coordinate for each species within each site
Almohadillas.df <- Almohadillas.df %>%
  group_by(Site) %>%
  mutate(SpeciesOrder = as.integer(row_number())) # Convert to integer
# Create the ggplot for the three overlapped abundance curves
plot_low <- ggplot(data = Almohadillas.df, aes(x = as.factor(SpeciesOrder), y =
relative_abundance, color = Site)) +
  geom_point(size = 2) +
  geom_line(aes(group = Site), linewidth = 0.7) +
  xlab("Species Rank") +
  ylab("(Pi)") +
  scale_x_discrete(breaks = unique(Almohadillas.df$SpeciesOrder)) + # Set
breaks to integers

```

```
scale_y_continuous(trans = 'log10', breaks = c(0.001, 0.01, 0.1, 1),
  labels = c(0.001, 0.01, 0.1, 1),
  limits = c(0.001, 1)) +
  scale_color_manual(values = c("#E41A1C", "#377EB8", "#4DAF4A")) + #
Change color scheme
  theme_pubclean()
# Display the plot
print(plot_low)
```