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# MULTIVARIATE ANALYSIS OF ECUADORIAN PROVINCES AND PROTECTED AREAS BASED ON THE PRESENCE OF POISON DART FROGS (*dendrobatidae*) AND SOME INSIGHTS FOR THEIR CONSERVATION

# ANÁLISIS MULTIVARIADO DE LAS PROVINCIAS Y ÁREAS PROTEGIDAS DE ECUADOR BASADO EN LA PRESENCIA DE RANAS DARDO VENENOSAS (DENDROBATIDAE) Y CONSIDERACIONES PARA SU CONSERVACIÓN

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#### Abstract

Understanding the distribution patterns of species is crucial for developing effective conservation strategies, particularly in regions with high biodiversity and endemism like Ecuador. Amphibians, especially poison dart frogs (Dendrobatidae), serve as important bioindicators of environmental health, yet they face significant threats from habitat loss, climate change, and other anthropogenic factors. This study examines the distribution patterns of poison dart frog species across various provinces and protected areas in Ecuador, utilizing an updated database containing 48 species, 32 of which are endemic to the country. Cluster Analysis and Principal Component Analysis (PCA) were applied to compare provinces and protected areas based on species richness, effectively identifying regions with higher and lower poison dart frog species presence. Additionally, ecological factors and the influence of protected areas on the distribution of these frogs are discussed. The findings reveal regions of high species richness, underscore the potential effects of environmental changes on poison dart frog communities, and highlight the crucial role of protected areas in safeguarding biodiversity. This study underscores the importance of integrating these analyses into conservation planning and decision-making processes, aiming to protect poison dart frogs and their habitats. By addressing these challenges, this research contributes complementary perspectives into preserving Ecuador's unique amphibian diversity.

*Keywords*: Poison dart frogs, *Dendrobatidae*, multivariate analysis, biodiversity, Ecuadorian provinces, Ecuadorian protected areas.

#### Resumen

La comprensión de los patrones de distribución de las especies es fundamental para desarrollar estrategias de conservación efectivas, especialmente en regiones con alta biodiversidad y endemismo como Ecuador. Los anfibios, y en particular las ranas venenosas (Dendrobatidae), son importantes bioindicadores de la salud ambiental, pero enfrentan amenazas significativas como la pérdida de hábitat, el cambio climático y otros factores antropogénicos. Este estudio analiza los patrones de distribución de las especies de ranas venenosas en varias provincias y áreas protegidas de Ecuador, utilizando una base de datos actualizada que incluye 48 especies, 32 de las cuales son endémicas del país. Se emplearon técnicas de Análisis de Clasificación (Cluster analysis) y de Ordenamiento (Análisis de componentes principales) para comparar provincias y áreas protegidas en función de su riqueza de especies, identificando regiones con mayor y menor presencia de estas ranas venenosas. Además, se discuten los factores ecológicos y la influencia de las áreas protegidas en la distribución de estas especies. Los hallazgos revelan regiones con alta riqueza de especies, resaltan los posibles efectos de los cambios ambientales en las comunidades de ranas venenosas y subrayan el papel crucial de las áreas protegidas en la preservación de la biodiversidad. El presente estudio enfatiza la importancia de integrar estos análisis en la planificación de conservación y en los procesos de toma de decisiones, contribuyendo a la protección de las ranas venenosas y de sus hábitats, así como a la preservación de la singular diversidad de anfibios en Ecuador.

*Palabras clave*: Ranas dardo venenosas, *Dendrobatidae*, análisis multivariado, biodiversidad, provincias ecuatorianas, áreas protegidas de Ecuador.

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# 1 Introduction

Ecuador is an exceptionally diverse country, home to a wide variety of habitats and niches that support numerous vertebrate and invertebrate species. As of 2023, 676 amphibian species have been reported in Ecuador, of which 324 are endemic (Ron and Ortiz, 2024). The Dendrobatidae family, commonly known as the *poison dart frog* family, comprises neotropical frogs characterized by their conspicuous coloration (Figures 1-4), which is associated with toxic alkaloids present on their skin (Daly et al., 2005; Patocka et al., 1999).



Figure 1. A specimen of *Ameerega bilinguis* (Dendrobatidae) (Gallice, 2009).

dart frogs (Daly et al., 1985, 2005; Spande et al., 1992). Species from the genus *Phyllobates* produce batrachotoxins, among the most potent non-peptide toxins, primarily targeting cardiac and nerve tissue by inducing irreversible membrane depolarization and disrupting normal signaling (Patocka et al., 1999). Conversely, species with lower amounts of skin toxins tend to exhibit duller or cryptic coloration (Santos et al., 2003).

According to a richness model created by Gómez (2017) the regions in Ecuador with highest Dendrobatidae species richness are the southeast and northwest amazonian region, the western foothills of the Ecuadorian Chocó and the eastern central Andean foothills. This shows a preference for humid and lowland areas, also reported by Valencia et al. (2009a,b).







Figure 2. A specimen of *Epipedobates anthonyii* (Dendrobatidae) (Tubifex, 2010).

Studying frogs from this family is relevant to numerous fields of research. Many alkaloids with potentially significant pharmacological properties have been isolated from the secretions of poison



Figure 4. A specimen of *Epipedobates machalilla* (Dendrobatidae) (Amphibiaweb, 2022).

The objectives of this research are: to analyze the distribution patterns of poison dart frog species across various provinces and protected areas in Ecuador, utilizing multivariate statistical methods such as Agglomerative Cluster Analysis and Principal Component Analysis to identify areas (provinces or protected areas) with high species richness and similarities between them. Also, to evaluate the influence of varying ecological conditions, as well as the role of protected areas on the presence, distribution, and conservation of poison dart frog species in Ecuador.

# 2 Materials and methods

#### 2.1 Study Area

This study focuses on 23 provinces (Figure 5; Appendix 1) and 18 national protected areas (Figure 6; Appendix 2) across mainland Ecuador. The analysis covers a wide array of habitats, including lowland tropical rainforests and montane cloud forests, which are typical environments for poison dart frogs.

The provinces addressed in the study include Coastal provinces, Western Andean-Coastal, Eastern Andean-Amazonian, and Amazonian provinces. The protected areas included exclusively national parks and reserves that are part of the National System of Protected Areas (SNAP) (Ministerio del Ambiente, Agua y Transición Ecológica, 2023), which play a crucial official role in conserving amphibian biodiversity.

#### 2.2 Data collection and processing

We considered a database consisting of the presence-absence of 48 Dendrobatidae frog species (Appendix 3), including 32 endemics to Ecuador, in the provinces and protected areas mentioned above. It is worth noting that the database was based on information from BioWeb, a webpage designed by the Museum of Zoology QCAZ (Ron and Ortiz, 2024). This website provides extensive information on amphibians in Ecuador, organized by families and species, as well as their geographical data.



Figure 5. Provinces of mainland Ecuador: Detailed information about them available in Appendix 1.



Figure 6. Protected Areas of mainland Ecuador belonging to the SNAP (National System of Protected Areas): Detailed information about them available in Appendix 2.

#### 2.2.1 Cluster analysis

To achieve comparative analysis of the different provinces and SNAP protected areas of Ecuador depending on their similarity (based on the presence/absence of Dendrobatidae species) an Agglomerative Cluster Analysis was applied (using Community Analysis Package 4.0, Pisces Conservation Ltd., 2014).

The Agglomerative Cluster Analysis, in this case using the Bray-Curtis index as a measure of dissimilarity, is a statistical method employed to group elements or samples (provinces or protected areas). In this technique, this index could be used to quantify the difference between pairs of samples, focusing on the presence of dendrobatid species.

The analysis begins by treating each sample (a province or a protected area) as its own cluster and then iteratively merges the closest clusters with another sample until a predefined number of clusters or a suitable clustering solution is achieved. This method is particularly useful in ecological studies for classifying sites, communities or habitats by their species composition, revealing patterns of biodiversity and community structure (Legendre and Legendre, 2012; Yánez and Quishpe, 2013).

Agglomerative clustering techniques are widely used in ecology to create dendrograms that illustrate the formation of clusters among elements of interest, such as provinces or protected areas. These dendrograms visually represent how these elements resemble one another while differing from others, providing valuable findings into their relationships and patterns (Clarke et al., 2016; Kassambara, 2017).

This technique was used in the study to evaluate groups of provinces or protected areas that exhibit similarities in their frog species richness.

#### 2.2.2 Principal component analysis

Principal component analysis (PCA) is a useful model that extracts the main patterns of a data set, considering variables and characteristics called principal components which are combined linearly in order to explain the variance of all variables (Greenacre et al., 2022; Wold et al., 1987).

PCA is also a statistical ordination technique used to simplify the complexity of highdimensional data while retaining its essential patterns and relationships. It does this by transforming the original variables into a new set of uncorrelated variables called principal components, which are ordered so that the first few retain most of the variation present in the original variables. PCA helps in identifying the underlying structure of the data, reducing dimensionality, and highlighting important relationships between variables. It is a statistical technique that effectively summarize the essential information in the data (Jolliffe and Cadima, 2016) and it is widely used in various fields such as biology, ecology, and social sciences to make data analysis more manageable and insightful (Abdi and Williams, 2010; Jolliffe, 2002; Molina et al., 2018).

PCA was employed in this study (using Community Analysis Package 4.0, Pisces Conservation Ltd., 2014) to explore the relationships among dendrobatid species, as well as their associations with the provinces or protected areas of Ecuador. These relationships were visualized through two ordination plots, providing a clearer and more comprehensive representation of species distribution and their corresponding locations.

# **3** Results and Discussion

#### 3.1 Similarity among provinces based on Dendrobatid species richness

The overall species richness by province is listed in Appendix 1. Notably, Morona Santiago and Napo (14 species each), Pastaza and Cotopaxi (13 each), Sucumbíos (11), and Santo Domingo and Pichincha (10 each) are the provinces where 10 or more species have been recorded.

The dendrogram displayed (Figure 7) illustrates the hierarchical clustering of Ecuadorian provinces based on the similarity of their dendrobatid species composition. Each individual branch represents a province, and the length of the branches reflects the level of dissimilarity between clusters. The horizontal axis represents a dissimilarity scale (Bray-Curtis index) ranging from 0 (complete similarity) to 1 (maximum dissimilarity). Provinces that form clusters at shorter branch lengths share more similar species compositions, indicating overlapping ecological characteristics or shared habitat types.

The ordination plot displayed (based on a Principal Components Analysis-Covariance Matrix; Figure 8) illustrates the relationships between dendrobatid species (green vectors) and the provinces of Ecuador (red squares). Provinces that are together share more similar species compositions, also indicating overlapping ecological characteristics or shared habitat types.

The analysis of dendrobatid species distributions across Ecuador's provinces reveals notable patterns of similarity, clustering provinces into four distinct groups (Figures 7-8). These groups illustrate how geographic, climatic, and ecological factors shape species richness and composition, providing a basis for understanding biogeographical trends and conservation needs.

#### Group 1: Azuay, Cañar, Bolívar, Chimborazo, Santa Elena, El Oro, Loja, and Guayas

This group is primarily characterized by species such as *Hyloxalus infraguttatus* (Hylinf), found in all eight provinces, *Hyloxalus elachyhistus* (Hylela), and *Epipedobates machalilla* (Epimac), both present in five provinces. Additionally, *Epipedobates anthonyi* (Epiant) is present in four provinces. These species are supported by shared environmental conditions, including similar altitudes and ecosystems (at least in the regions where these frog species were historically found).

Other species, such as *Hyloxalus vertebralis* (Hylver) and *Hyloxalus jacobuspetersi* (Hyljac), occur in fewer provinces but still contribute to the group's biodiversity. These patterns align with findings by Santos and Cannatella (2011) and Gómez (2017), which highlight the role of habitat connectivity in fostering shared species richness

#### Group 2: Carchi, Imbabura, Santo Domingo, Cotopaxi, Pichincha, Esmeraldas, Manabí, and Los Ríos

Provinces in this group exhibit high similarity due to the presence of species like *Epipedobates boulenge-ri* (Epibou), present in all eight provinces, *Hyloxalus* 

*awa* (Hylawa) and *Oophaga sylvatica* (Oopsyl), both found in seven provinces, and *Epipedobates espinosai* (Epiesp) in six provinces. Additionally, *Epipedobates machalilla* (Epimac) and *Hyloxalus infraguttatus* (Hylinf) are present in five and four provinces, respectively.

The proximity of these provinces and their shared lowland tropical and subtropical forest habitats support a high degree of overlap in dendrobatid species, as noted in other studies on amphibian distribution in Ecuador (Jongsma et al., 2014; Gómez, 2017).

#### Group 3: Morona Santiago, Pastaza, Napo, Sucumbíos, and Orellana

This cluster, encompassing Amazonian provinces, is characterized by *Ranitomeya ventrimaculata* (Ranven), *Ameerega hahneli* (Amehah), *Ranitomeya variabilis* (Ranvar), and *Hyloxalus sauteri* (Hylsau), all present in the five provinces. *Ameerega bilinguis*  (Amebil) and *Ameerega parvula* (Amepar) appear in four provinces. The high species richness observed here reflects the continuous forest cover and the variety of ecological niches of the Amazon basin.

Studies by Myers et al. (2000) emphasize the Amazon's role as a biodiversity hotspot, underscoring the need to mitigate threats like deforestation and habitat fragmentation.

#### Group 4: Tungurahua and Zamora Chinchipe

This group is defined by *Hyloxalus shuar* (Hylshu), present in both provinces, alongside species such as *Hyloxalus anthracinus* (Hylant), *Hyloxalus marmoreoventris* (Hylmar), *Hyloxalus mystax* (Hylmys), *Hyloxalus exasperatus* (Hylexa), and *Leucostethus fugax* (Leufug), each found in one province. The distinct species composition suggests that altitudinal barriers and special microhabitats could play a pivotal role in shaping biodiversity in these areas.



Figure 7. Dendrogram of 23 provinces of mainland Ecuador based on the presence-absence of 48 species of dendrobatids frogs. Method used: Hierarchical Agglomerative, Complete Linkage, and Bray-Curtis dissimilarities between provinces or groups of provinces (top axis).

These patterns align with conclusions from Ortiz et al. (2013), which highlight the Andes as centers of endemic amphibian diversity.

Regarding the situation in these two provinces, they are physically separated and share few similar environments, particularly in the eastern region of Tungurahua and the western region of Zamora Chinchipe. This geographic and ecological separation likely contributes to the low number of shared dendrobatid species between them, such as *Hyloxalus shuar*. Consequently, the similarity between these two provinces is among the lowest observed (Figure 7).

#### Necessary considerations

The clustering of provinces reflects biogeographical and ecological drivers of dendrobatid species distributions. The Amazonian provinces form cohesive clusters due to their uniform environmental conditions, while Andean and coastal provinces exhibit more distinct groupings influenced by altitudinal gradients, climatic variability, and habitat specialization. The dendrogram patterns underscore the interplay between geographic barriers and ecological connectivity in shaping biodiversity, offering critical insights for targeted conservation planning.



Figure 8. Ordination Plot (based on a PCA using a Covariance matrix) of the 23 provinces of Ecuador and their dendrobatids species richness (48). Note: F1 (horizontal axis) absorbed 43.4% of the variance; F2 (vertical), 22.6%.

The Amazon plays a pivotal role in preserving Ecuador's dendrobatid diversity, emphasizing the urgent need to safeguard its habitats from deforestation and other anthropogenic pressures. In contrast, Andean provinces such as Tungurahua, Chimborazo, and Bolívar host fewer dendrobatid species due to more difficult climatic conditions and more fragmented habitats. Coastal provinces, with relatively continuous ecosystems, facilitate species dispersal and persistence, yet face growing threats from agricultural expansion and urbanization, underscoring the necessity of targeted conservation measures.

Patterns of similarity and dissimilarity among provinces reveal the influence of biogeographic barriers on species distribution. For example, despite their relative proximity, Zamora Chinchipe and Napo exhibit distinct species assemblages shaped by microhabitat and environmental differences. Tools like Bray-Curtis index highlight these disparities, providing insights into the ecological and evolutionary drivers of dendrobatid distributions.

These findings stress the importance of spatial analyses in conservation planning. Andean and coastal provinces with unique species compositions require focused conservation strategies, while the high similarity among Amazonian provinces highlights the need to maintain habitat connectivity. Ecuador's exceptional dendrobatid diversity underscores the nation's role as a global amphibian biodiversity hotspot and the urgency of conserving microhabitats to protect vulnerable populations from human-induced pressures.

# 3.2 Similarity among SNAP's protected areas based on Dendrobatid species richness

The overall species richness by SNAP protected area is listed in Appendix 2. Remarkably, Yasuní National Park and Ilinizas Ecological Reserve (7 species each), Sangay National Park and Cuyabeno Reserve (5 each), and non-SNAP areas (mainly in private conservation zones) (21). These four protected areas also host the highest number of endemic species.

The dendrogram in Figure 9 illustrates the hierarchical clustering of Ecuadorian protected areas within the SNAP system, based on the similarity of their dendrobatid species composition, and one branch representing the areas outside this system (F-SNAP).

Each branch corresponds to a protected area, with branch lengths representing the degree of dissimilarity between clusters. The horizontal axis depicts the Bray-Curtis dissimilarity index, ranging from 0 (complete similarity) to 1 (maximum dissimilarity). Clusters formed at shorter branch lengths indicate protected areas with more similar species compositions, likely reflecting overlapping ecological characteristics or shared habitat types.

The ordination plot displayed (based on a principal component analysis-covariance matrix; Figure 10) illustrates the relationships between dendrobatid species (green vectors) and the SNAP's protected areas (red squares). Protected areas that are together share more similar species compositions, also indicating overlapping ecological characteristics or shared habitat types or geographic proximity.

The analysis of dendrobatid species distributions across Ecuador's protected areas (AAPP) reveals distinct patterns of similarity, clustering areas into eight groups based on shared species compositions (Figures 9–10). These groupings highlight the role of geographic proximity, habitat connectivity, and ecological characteristics in shaping species richness and diversity, offering critical insights into biogeographical trends and conservation priorities.

#### Group 1: Machalilla and Manglares Churute

This cluster of coastal protected areas is defined exclusively by the presence of *Epipedobates machalilla* (Epimac), a species endemic to this region. Shared lowland ecosystems and coastal forest remnants contribute to the group's composition, although habitat loss from urbanization and agriculture poses significant threats.

#### Group 2: Chimborazo and Pasochoa

*Hyloxalus jacobuspetersi* (Hyljac) is the sole species found in both areas, marking this cluster. The distinct altitudinal and climatic conditions of these regions may limit species richness but provide critical refuges for specialized taxa.

#### Group 3: Yasuní, Cuyabeno, and Limoncocha

This cluster is defined by the presence of four species: *Ameerega bilinguis* (Amebil), *Ameerega hahneli* (Amehah), *Hyloxalus sauteri* (Hylsau), and *Ranitomeya variabilis* (Ranvar), all found in the three areas. These species benefit from the extensive continuous forest cover of the Amazon basin, which facilitates dispersal and gene flow. These findings align with Myers et al. (2000), emphasizing the Amazon's significance as a biodiversity hotspot.

Multivariate analysis of Ecuadorian provinces and protected areas based on the presence of poison dart frogs (Dendrobatidae) and some insights for their conservation



Figure 9. Dendrogram of 18 Protected Areas in Mainland Ecuador within the SNAP, Based on the Presence-Absence of 48 Dendrobatid Frog Species, and one general area for places outside the SNAP (F-SNAP). The analysis was conducted using the Hierarchical Agglomerative Method with Complete Linkage and Bray-Curtis dissimilarities (top axis).

#### Group 4: Mache-Chindul, Manglares Cayapas Mataje, Ilinizas, and La Chiquita

This group is characterized by *Epipedobates boulengeri* (Epibou), present in all four areas, alongside *Hyloxalus infraguttatus* (Hylinf) and *Oophaga sylvatica* (Oopsyl), found in three areas except La Chiquita. These species benefit from shared lowland tropical habitats and relatively high habitat connectivity across these protected areas.

#### Group 5: Sangay and Cajas

The shared presence of *Hyloxalus vertebralis* (Hylver) and *Hyloxalus anthracinus* (Hylant) characterizes this group. Both species are associated with mid-to relatively high-altitude habitats, highlighting the influence of Andean ecosystems on dendrobatid distributions. Similar altitudinal gradients and climatic conditions likely support the overlapping compositions observed.

# Group 6: Cayambe-Coca, Sumaco-Napo Galeras, and Antisana

This group is primarily distinguished by *Hyloxalus pulchellus* (Hylpul) and *Hyloxalus bocagei* (Hylboc), present across the three areas. These protected areas share montane forest habitats, characterized by steep slopes, high precipitation, and a diverse array of ecological niches, fostering species persistence.

#### **Group 7: Llanganates**

Uniquely represented by *Hyloxalus maculosus* (Hylmac), Llanganates forms a solitary group, reflecting its distinct ecological and geographical conditions. The species' restricted range underscores the importance of preserving isolated Andean habitats.



Figure 10. Ordination Plot (based on a PCA using a Covariance matrix) of the 18 Protected Areas of Mainland Ecuador (SNAP) and one general area for places outside the SNAP (F-SNAP) and their dendrobatids species richness (48). Note: F1 (horizontal) accounts for 31.5% of the variance; F2 (vertical) explains 20.2%.

#### **Group 8: Podocarpus**

Podocarpus stands alone, hosting *Hyloxalus cevallosi* (Hylcev), *Hyloxalus mystax* (Hylmys), and *Leucostethus fugax* (Leufug). This protected area shares *Hyloxalus breviquartus* (Hylbrev) with areas outside SNAP (F-SNAP). This species composition reflects the ecological complexity of Podocarpus, characterized by its relatively high-altitude cloud forests and endemic-rich flora and fauna.

The Podocarpus National Park joins very late in the Cladogram with the entity outside the SNAP (F-SNAP).

Beyond the SNAP system (F-SNAP), 21 dendrobatid species have been exclusively recorded in smaller conservation areas or wild environments outside formal national protection schemes. These include privately managed reserves (e.g., Bosque Protector Mindo Nambillo, Maquipucuna Reserve, Tesoro Escondido Reserve, Bilsa Biological Station, Río Palenque Research Center) and municipal or provincial conservation areas. These species are: *Epipedobates anthonyi, E. darwinwallacei, E. espino*- sai, E. tricolor, Excidobates captivus, E. condor, Hyloxalus delatorreae, H. elachyhistus, H. exasperatus, H. fallax, H. italoi, H. maquipucuna, H. marmoreoventris, H. peculiaris, H. pumilus, H. shuar, H. toachi, Leucostethus bilsa, Paruwrobates erythromos, P. whymperi, Ranitomeya reticulata, all of them inhabiting these not-SNAP protected areas, emphasizing the importance of broader conservation strategies beyond the SNAP system.

#### **Necessary considerations**

The clustering of Ecuador's protected areas based on dendrobatid species composition reveals how ecological, geographic, and biogeographical factors influence biodiversity patterns. Amazonian areas exhibit high similarity due to their continuous forest cover, while Andean and coastal regions show distinct groupings shaped by altitudinal gradients, habitat specialization, and climatic variability.

The high level of dissimilarity among certain areas (or groups of areas) highlights the need for a networked approach to conservation. Methods such as network theory have proven effective for

identifying and delimiting biogeographical regions, enabling more targeted and efficient conservation strategies (Vilhena and Antonelli, 2015). Each area contributes uniquely to the overall biodiversity, and protecting a diverse range of habitats is crucial as many species with restricted distributions depend on specific environmental conditions for their survival (Guisan et al., 2013).

These findings underscore the urgent need for conservation actions tailored to preserve Ecuador's exceptional amphibian diversity and address threats such as deforestation, habitat fragmentation, and human-induced pressures.

#### 3.3 Distribution patterns and conservation implications for poison dart frogs included in threatened species categories

According to the information on Threatened Categories provided by the International Union for Conservation of Nature (International Union for Conservation of Nature, 2009) (Appendix 3), the following are the poison dart frog species classified under these categories:

#### 3.3.1 Species in category Vulnerable

The following species are categorized as vulnerable: *Epipedobates espinosai, Hyloxalus infraguttatus, H. ne-xipus, H. peculiaris, H. pulchellus, H. shuar, H. toachi,* and *H. vertebralis.* These frogs are at risk of population decline due to factors such as habitat loss, environmental degradation, and climate change.

#### 3.3.2 Species in category Endangered

The following species are categorized as endangered: *Ectopoglossus confusus, Epipedobates tricolor, Excidobates captivus, E. condor, Hyloxalus breviquartus, H. elachyhistus, H. fuliginosus, H. lehmanni, H. maculosus, H. marmoreoventris, H. mystax, Paruwrobates erythromos,* and *P. whymperi.* These species face severe risks of extinction in the wild if immediate conservation actions are not implemented. Habitat destruction and human activities are the primary drivers of their declining populations.

#### 3.3.3 Species in category Critically Endangered

These species are at an extremely high risk of extinction due to their limited distributions, fragmented

populations, and ongoing habitat destruction. Conservation efforts must urgently focus on their survival. The Critically Endangered species include *Andinobates abditus*, *Hyloxalus anthracinus*, *H. bocagei*, *H. delatorreae*, *H. exasperatus*, *H. fallax*, *H. jacobuspetersi*, *H. maquipucuna*, *H. marmoreoventris*, and *Hyloxalus pumilus*.

# 3.3.4 Importance of provincial and protected area data

Understanding the distribution of these species across provinces and protected areas is critical for identifying biodiversity hotspots and prioritizing conservation efforts.

In Ecuador, the hotspots for dendrobatid frogs are primarily located in these three regions (Tapia et al., 2017; Centro Jambatu de Investigación y Conservación de Anfibios, 2024):

- The Amazon Basin (eastern Ecuador): specially the provinces of Napo, Morona Santiago, and Sucumbíos, are known for their general high biodiversity. The dense rainforests and wetlands in this area provide the moisture and microhabitats needed for these frogs to thrive.
- The Chocó Bioregion (western Ecuador): specially the Esmeraldas and Pichincha provinces host many dendrobatid species thanks to the humid forests and diverse altitudinal gradients.
- The eastern Andean slopes, including Zamora-Chinchipe, Morona Santiago, and Azuay, also harbor dendrobatid, especially in cloud forests at varying altitudes.

Each province has unique ecological characteristics that influence the presence and survival of these frogs and other important species (Delgado et al., 2023; Crespo et al., 2022). By mapping their distribution, conservationists can pinpoint regions with the highest biodiversity or those with the most threatened populations, ensuring resources are allocated effectively. For example, provinces with high concentrations of Endangered or Critically Endangered species may require stricter habitat protection laws, targeted restoration projects, or the establishment of ecological corridors to maintain connectivity between populations.

#### 4 Conclusions implications

This study highlights the significant biodiversity of Ecuador's poison dart frogs, with distinct distribution patterns influenced by geographic, climatic, and ecological factors. The clustering of provinces and protected areas into groups based on species composition reveals clear biogeographic patterns, with Amazonian regions displaying high species richness and similarity. In contrast, Andean and coastal areas exhibit more unique species assemblages, shaped by altitudinal gradients and varying habitat conditions. These findings underscore the need for region-specific conservation efforts tailored to the unique environmental characteristics of each area or species composition.

The clustering of provinces also illustrates how the integrity of ecosystems is essential for maintaining amphibian biodiversity. In particular, the Amazonian provinces, which share continuous forest cover, support a greater diversity of species. In contrast, Andean provinces, which face harsher environmental conditions, show fewer species, emphasizing the vulnerability of these high-altitude ecosystems. The results suggest that protecting continuous forest habitats in lowland regions is critical to conserving Ecuador's amphibian diversity.

Additionally, the role of Ecuador's National System of Protected Areas (SNAP) in safeguarding biodiversity is evident. Protected areas such as Yasuní, Cuyabeno, and Sangay harbor a wide variety of species, demonstrating the importance of these areas in maintaining species richness. However, the dissimilarity observed among some SNAP areas indicates that some regions may require more targeted management strategies to address their unique ecological challenges. The identification of areas with lower species richness highlights the need for conservation actions that focus on habitat restoration, preventing further fragmentation.

While SNAP areas play a key role in biodiversity conservation, the study also reveals the importance of extending national conservation efforts beyond these formal protected spaces. Smaller reserves and privately managed areas, such as the Bosque Protector Mindo Nambillo and Maquipucuna Reserve, also host significant dendrobatid species. These areas,

and Management while outside the SNAP, are critical for maintaining connectivity and preserving species with limited distributions generally categorized as Threatened Species. Thus, a more integrated conservation approach, including private and community-led efforts, is essential for the long-term sustainability of Ecuador's amphibian populations.

> The study's findings provide a comprehensive understanding of the distribution patterns of Ecuador's dendrobatid species and the ecological factors shaping them. Conservation planning should prioritize the preservation of continuous habitats, connectivity between protected areas, the presence of Dart Frog Threatened Species and the protection of specialized ecosystems, especially in Andean and coastal regions. A networked conservation approach, combining the efforts of SNAP and smaller conservation initiatives, is crucial to ensuring the future of Ecuador's exceptional amphibian biodiversity. Effective management strategies should focus on both the protection of existing habitats and the restoration of fragmented areas to prevent further biodiversity loss.

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# Author's contribution

P.Y.M.: Conceptualization, Data curation, Formal analysis, Funding acquisition, Investigation, Methodology, Project administration, Computing resources, Supervision, Validation, Visualization, Writing– original draft, Writing– review editing. J.G.G.: Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Validation, Writing- original draft, Writing- review editing. A.H.E., M.B.M., and D.Q.S. performed the Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Validation, and Writing- original draft.

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# Appendix

# Appendix 1

No.	Province	Region	Province Acronym used in multivariate analysis	Species richness of dendrobatid per province
1	Azuay	Andean	AZU	8
2	Bolívar	Andean	BOL	7
3	Cañar	Andean	CAÑ	5
4	Carchi	Andean	CAR	7
5	Chimborazo	Andean	CHI	1
6	Cotopaxi	Andean	COT	13
7	El Oro	Coastal and Western Andean	ORO	3
8	Esmeraldas	Coastal and Western Andean	ESM	9
9	Guayas	Coastal and Western Andean	GUA	4
10	Imbabura	Andean	IMB	7
11	Loja	Andean	LOJ	4
12	Los Ríos	Western Andean	RIOS	6
13	Manabí	Coastal and Western Andean	MANB	7
14	Morona Santiago	Amazonian and Eastern Andean	MORSAN	14
15	Napo	Amazonian and Eastern Andean	NAP	14
16	Orellana	Amazonian	ORE	9
17	Pastaza	Amazonian and Eastern Andean	PAST	13
18	Pichincha	Andean	PICH	10
19	Santa Elena	Coastal	SELEN	2
20	Santo Domingo de los Tsáchilas	Western Andean	SDOM	10
21	Sucumbíos	Amazonian and Eastern Andean	SUC	11
22	Tungurahua	Andean	TUNG	3
23	Zamora Chinchipe	Amazonian and Eastern Andean	ZAMCH	8

Table 1. Provinces of mainland Ecuador addressed in the study

# Appendix 2

	Category of	Protected Area	Region	Province Acronym	Species
No.				used in multivariate	richness of dendrobatid
	T Touceur Area			analysis	per area
1	Parque Nacional	PN Cajas	Andean	CAJAS	2
2	Parque Nacional	PN Cayambe Coca	Amazonian and Eastern Andean	CAY-COC	3
3	Parque Nacional	PN Llanganates	Amaznoian and Eastern Andean	LLANG	1
4	Parque Nacional	PN Machalilla	Coastal	MACHAL	1
5	Parque Nacional	PN Podocarpus	Amazonian and Eastern Andean	PODOC	4
6	Parque Nacional	PN Sangay	Amazonian and Eastern Andean	SANGAY	5
7	Parque Nacional	PN Sumaco Napo Galeras	Amazonian and Eastern Andean	SUMACO NG	3
8	Parque Nacional	PN Yasuní	Amazonian	YASUN	7
9	Reserva de Producción Faunística	RPF Chimborazo	Andean	CHIMB	1
10	Reserva de Producción Faunística	RPF Cuyabeno	Amazonian	CUYAB	5
11	Reserva Biológica	RB Limoncocha	Amazonian	LIMONC	4
12	Reserva Ecológica	RE Antisana	Eastern Andean	ANTIS	1
13	Reserva Ecológica	RE Ilinizas	Andean	ILINIZ	7
14	Reserva Ecológica	RE Mache Chindul	Coastal	MACHE	4
15	Reserva Ecológica	RE Manglares Cayapas Mataje	Coastal	MANG-CAYPS	3
16	Reserva Ecológica	RE Manglares Churute	Coastal	MANG-CHURU	1
17	Refugio de Vida Silvestre	RVS Pasochoa	Andean	PASOCH	1
18	Refugio de Vida Silvestre	RVS La Chiquita	Coastal	CHIQUIT	1
_	Only outside SNAP protected areas *	_	Various	F-SNAP	21

Table 2. SNAP National Protected Areas addressed in the study

\* Although some species of dendrobatid frogs were recorded outside the protected areas of the National System of Protected Areas (SNAP), it is worth mentioning that most of them were recorded in smaller private, provincial, or municipal conservation areas, such as protective forests or similar areas

# Appendix 3

Species Acronym					Concernation
No.	Species	used in Multivariate Analysis	Common name	relation to Ecuador	Conservation Category (IUCN, 2024)
1	Ameerega bilinguis	Amebil	Ecuador poison frog	Endemic	Least Concern
2	Ameerega hahneli	Amehah	Yurimaguas poison frog	Native	Least Concern
3	Ameerega parvula	Amepar	Valle Santiago rocket frog	Native	Least Concern
4	Andinobates abditus	Andabd	Collins' poison frog	Endemic	Critically Endangered
5	Ectopoglossus confusus	Ectcon	Confusing Rocket Frog	Endemic	Endangered
6	Epipedobates anthonyi	Epiant	Epibatidine poison frog	Native	Near Threatened
7	Epipedobates boulengeri	Epibou	Marbled poison-arrow frog	Native	Least Concern
8	Epipedobates darwinwallacei	Epidar	Darwin and Wallace poison frog	n.d.	n.d.
9	Epipedobates espinosai	Epiesp	Espinosa poison frog	Endemic	Vulnerable
10	Epipedobates machalilla	Epimac	Machalilla rocket frog	Endemic	Least Concern
11	Epipedobates tricolor	Epitri	Tricolor poison frog	Endemic	Endangered
12	Excidobates captivus	Exccap	Río Santiago poison frog	Native	Endangered
13	Excidobates condor	Exccon	Cóndor Poison Frog	Endemic	Endangered
14	Hyloxalus anthracinus	Hylant	South american rocket frog	Endemic	Critically Endangered
15	Hyloxalus awa	Hylawa	Awa rocket frog	Endemic	Near Threatened
16	Hyloxalus bocagei	Hylboc	Bocage's rocket frog	Endemic	Critically Endangered
17	Hyloxalus breviquartus	Hylbrev	Urrao rocket frog	Native	Endangered
18	Hyloxalus cevallosi	Hylcev	Palanda rocket frog	Endemic	Near Threatened
19	Hyloxalus delatorreae	Hyldel	Stella Rocket Frog	Endemic	Critically Endangered
20	Hyloxalus elachyhistus	Hylela	Loja rocket frog	Native	Endangered
21	Hyloxalus exasperatus	Hylexa	Yapitya rocket frog	Endemic	Critically Endangered
22	Hyloxalus fallax	Hylfal	Cotopaxi rocket frog	Endemic	Critically Endangered
23	Hyloxalus fuliginosus	Hylful	Quijos rocket frog	Endemic	Endangered

 Table 3. Dendrobatidae frog species included in the study

24	Hyloxalus infraguttatus	Hylinf	Chimbo rocket frog	Endemic	Vulnerable
25	Hyloxalus italoi	Hylita	Pastaza rocket frog	Endemic	Near Threatened
26	Hyloxalus jacobuspetersi	Hyljac	Quito rocket frog	Endemic	Critically Endangered
27	Hyloxalus lehmanni	Hylleh	Lehmann's rocket frog	Native	Endangered
28	Hyloxalus maculosus	Hylmac	Spotted rocket frog	Endemic	Endangered
29	Hyloxalus maquipucuna	Hylmaq	Maquipucuna rocket frog	Endemic	Critically Endangered
30	Hyloxalus marmoreoventris	Hylmar	Río Negro rocket frog	Endemic	Critically Endangered
31	Hyloxalus mystax	Hylmys	Cloud forest rocket frog	Endemic	Endangered
32	Hyloxalus nexipus	Hylnex	Los Tayos rocket frog	Native	Vulnerable
33	Hyloxalus peculiaris	Hylpec	Funny rocket frog	Endemic	Vulnerable
34	Hyloxalus pulchellus	Hylpul	Espada's rocket frog	Native	Vulnerable
35	Hyloxalus pumilus	Hylpum	San Vicente rocket frog	Endemic	Critically Endangered
36	Hyloxalus sauli	Hylsau	Santa Cecilia rocket frog	Native	Near Threatened
37	Hyloxalus shuar	Hylshu	Shuar rocket frog	Endemic	Vulnerable
38	Hyloxalus toachi	Hyltoa	Toachi rocket frog	Endemic	Vulnerable
39	Hyloxalus vertebralis	Hylver	Cuenca's rocket frog	Endemic	Vulnerable
40	Hyloxalus yasuni	Hylyas	Yasuní rocket frog	Endemic	Near Threatened
41	Leucostethus bilsa	Leubil	Bilsa white-chested frog	Endemic	Not Evaluated
42	Leucostethus fugax	Leufug	Pastaza rocket frog	Endemic	Near Threatened
43	Oophaga sylvatica	Oopsyl	Little-devil poison frog	Native	Near Threatened
44	Paruwrobates erythromos	Parery	Palenque poison frog	Endemic	Endangered
45	Paruwrobates whymperi	Parwhy	Tanti rocket frog	Endemic	Endangered
46	Ranitomeya reticulata	Ranret	Red-backed poison frog	Native	Near Threatened
47	Ranitomeya variabilis	Ranvar	Yellow striped poison frog	Native	Least Concern
48	Ranitomeya ventrimaculata	Ranven	Amazonian Poison Frog	Native	Least Concern