

Trends of amphibian species discovery: an overview of the geographical and social factors affecting amphibian taxonomy between 2000-2019

The Red-eyed Tree Frog **(Agalychnis callidryas)** *Image: (Bildagentur Zoonar, 2015)*

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Contents

Abstract

Taxonomy – the science of classifying and naming organisms – is facing challenges due to reduced funding and positions available. Taxonomy and conservation are also interlinked, making taxonomy increasingly important in the future. The effects of the current state of taxonomy has not been explored in relation to a specific taxon before, so in this metaanalysis, the trends in species discovery were explored within the class Amphibia, as well as within the researchers, funders and journals publishing this research. Using new species data from *Amphibian Species of the World*, this investigation found that between 2000- 2019, most newly discovered amphibians were found in Asia and South America, with an average of 114 species described per year. New species are being discovered in protected areas 38.62% of the time, and traits such as decreasing body size, exhibited in recently discovered frogs, suggest they may be increasingly difficult to find in the future. Most researchers discovering amphibians originate from the United States and Brazil. The United States was also the most important funder of taxonomic research. Approximately 42% of published research on new amphibians is open access, and only 12.9% of published research on new species have genetic data available for them. As amphibians are threatened by anthropogenic climate change, and are still undergoing high levels of discovery, effort to ensure funding and resources continue to be available to taxonomists is of paramount importance to inform conservation efforts, and keep the knowledge of Earth's biodiversity current and accurate.

Abbreviations

- ASW Amphibian Species of the World (Frost, 2024)
- eDNA Environmental DNA
- GDP Gross Domestic Product (per capita)
- GLM General Linear Model
- GLMM Generalised Linear Mixed Model
- IUCN International Union for Conservation of Nature
	- LRT Likelihood Ratio Test
	- SVL Snout to vent length
	- VIF Variance Inflation Factor

Introduction

The current state of taxonomy

The title of Mertl's (2002) article "Taxonomy in danger of extinction" reflects a wellestablished and widespread sentiment that taxonomy is a dying discipline (Drew, 2011; Mertl, 2002). If true, this could have serious consequences on further acquisition of knowledge about the natural world (Wheeler, 2014). Unfortunately, taxonomy in its traditional form, where taxonomists identify and classify a species, but also analyse its evolutionary relationships to other species, and study the origins of life and its diversification, does appear to be on the decline (Bebber et al., 2014; Wheeler, 2014). Technological changes in the field, including the introduction of genetic techniques such as DNA barcoding and phylogenetic analyses, has allowed more execution of components of taxonomic work, namely species classification, leading to criticism that true holistic taxonomy is no longer being practised (Wheeler, 2014). Certain taxonomists, such as Wheeler (2014) have argued that these technological innovations have led to an increase in people dabbling in taxonomy, and publishing papers about new species, after only carrying out basic DNA analysis. This can result in errors including misidentification and mislabelling, leading to incorrect information about species numbers or evolutionary relationships (Drew,

2011; Löbl et al., 2023). Furthermore, due to increasing author credit on published taxonomic papers, misconception follows that there are increasing numbers of taxonomists, despite ongoing loss of funding and available positions (Bebber et al., 2014; Löbl et al., 2023; Wheeler, 2014). It has been shown that species-focused conservation is interlinked with taxonomy, as well as other fields such as disease ecology, and even forestry and farming, being informed and relying on species information provided by taxonomists (Löbl et al., 2023; Mace et al., 2004; Tapley et al., 2018). There are an estimated 8.67 million eukaryotic species existing today, with 86% of them awaiting discovery and/or description (Mora et al., 2011). Biodiversity is being lost at unprecedented rates despite new species still being discovered. Extinction rates are 100-1000 times greater than pre-human baseline levels (Pimm et al., 1995). With increasing loss of species each year, new species being discovered, and a lack of resources to declare and describe them, there is the danger of losing species before even learning of their existence (González-Oreja, 2008; Mora et al., 2011). Investment to learn about the range of biodiversity on our planet, what is being lost as well as to learn more about species interactions and ecosystem structure is necessary to correctly manage surviving populations (Löbl et al., 2023).

Other than the importance of taxonomy for furthering scientific understanding and aiding large scale industries, public interest in nature and the environment also deserves recognition (Manfredo, 1989; Mittermeier et al., 2021). Specific tools such as Wikipedia and iNaturalist allow the general public to educate themselves and engage with the natural world (Mittermeier et al., 2021; Ueda et al., 2008; Wikimedia, 2024a). Additionally, through citizen science, people can carry out surveys of their local biodiversity, enabling large scale data collection, and up to date information on the presence and abundance of species (Theobald et al., 2015). One such initiative, the BioBlitz, created by the National Biodiversity

Network, encourages scientist and general public volunteers to observe and record as many plant, animal and fungi species as possible in their local area during a 24-hour period (NBN, 2024). Peter et al. (2021) argue that educating the public on the importance of the natural world will help to ensure its protection in the future. In 2015, the publisher Elsevier began collaborating with Wikimedia Foundation to increase the accessibility of peer-reviewed published scientific articles to specific editors of Wikipedia. This ensures Wikipedia's information, one of the most heavily shared information sources online, is correct and up to date (Goodschild van Hilten, 2022). Accredited editors of Wikipedia may also be granted special access to specific journals through *The Wikipedia Library*, run by Wikimedia (2024b). Elsevier itself has been criticised in the past due its lack of open access papers, and for increasing costs to access academic journals (Schiermeier & Mega, 2017). While open access is beneficial for the sharing of knowledge, substantial article processing charges and publishing fees that authors need to pay may actually work against resource-poor fields (Frank et al., 2023).

While much research exists on the apparent dire state of taxonomy in general, little information exists about how this affects particular taxa. Understanding how the discovery and description of a specific group is impacted by today's state of taxonomy can provide valuable insights for conservation and future planning. This understanding includes being informed about how many species are being discovered, where this discovery is taking place, whether research is done within protected areas, as well as where those funding or carrying out the research are based. Additionally, investigation about whether the recently described species show any characteristics of taxonomic interest, or if DNA data is available for newly described species, and whether the papers published describing these species are open access would all be beneficial. To address these questions, a model group should be

chosen that has a high rate of discovery, is species-rich, and is globally distributed. An extensive amount of data should be available to analyse and ideally, a fairly comparable body plan within the group would be helpful for investigating trends in their characteristics. One group that fits all these criteria is the amphibians.

Figure 1: The three orders of amphibians (a) a yellow-banded poison dart frog (Dendrobates leucomelas) from the order Anura, (b) a fire salamander (Salamandra salamandra) *from the order Urodela, (c) a Kodagu striped Ichthyophis* (Ichthyophis Kodaguensis) *from the order Gymnophiona. Images: a (Dwyer-Lindgren, 2023), b (Milošević, 2023), c (Pal, 2011).*

Key features of amphibians

Amphibians are a class of vertebrates thought to represent the remnants of the first land vertebrates (Wake & Koo, 2018). The extant amphibians, lissamphibians, can be divided into 3 groups: frogs and toads (order Anura), salamanders and newts (order Urodela), and caecilians (order Gymnophiona) (Fig. 1) (Catenazzi, 2015; Wells, 2007). Anura is the largest group with approximately 7000 species, composing 88% of all amphibia (Wake & Koo, 2018). Anurans are particularly sensitive to habitat changes, due to their unique behaviour and physiology, and are often considered biological indicators, used as informants for ecosystem health (do Amaral et al., 2019). They are adapted to perform cutaneous respiration, carrying out part of their gas exchange through their thin integument (Wake & Koo, 2018). This means they often need to live in environments with a consistent temperature range and maintain access to water sources to prevent desiccation (Wells,

2007). It has been suggested that access to water also determines habitat selection, distribution of species, and reproductive success. Most anurans have a juvenile aquatic tadpole life-stage before undergoing metamorphosis to their terrestrial adult form. They therefore require multiple different habitat types throughout their lifecycle (Wells, 2007).

Amphibians as a model group

Amphibians are an ideal model group for many reasons. Firstly, they are widespread, with anurans being found on every continent except Antarctica and being concentrated in tropical regions (Wake & Koo, 2018). South America is the continent with the highest number of amphibians, and Brazil has the highest occurrence and rate of description of amphibian species in the world (Guerra et al., 2020; Segalla et al., 2019; Vasconcelos et al., 2019). Amphibians are also abundant on islands, such as Madagascar, the Philippines, Borneo, and New Guinea (Wake & Koo, 2018). Islands are particularly important due to their geographic isolation and high adaptive pressures, producing greater numbers of endemic species than the mainland (Clores et al., 2021; Kier et al., 2009; Siler et al., 2010).

Secondly, frog species are being discovered at high rates, and this shows no evidence of slowing down (Tapley et al., 2018). A decade ago, 25% of discovered amphibians up to that time had been found between 2005 and 2014 (Catenazzi, 2015). The increase in the rate of discovery is a result of both increased surveying capabilities, such as the use of bioacoustics to find and identify frogs, but also the development of genomic techniques to identify new species, as well as recognise multiple genetically distinct species previously thought to be a singular species (Kohler et al., 2005). Thirdly, anurans have a highly conserved body plan, useful for analysing changes in the physical traits exhibited over time (Wake & Koo, 2018).

Finally, there is a vast amount of data available on amphibians by virtue of the online Amphibian Species of the World (ASW) database. It is the official CITES (Convention on International Trade in Endangered Species of Wild Fauna and Flora) classification publication for amphibians, and is regularly updated, linking every new species described to its correct place within its clade, and linking the associated research papers (Frost, 2024).

Threats facing amphibians

Despite the high rates of new species discovery, many amphibians are at an elevated risk for extinction, making conservation a priority (IUCN, 2024; Kohler et al., 2005). Between 2004- 2024, the assessment of amphibians by the International Union for Conservation of Nature (IUCN) changed from 42% of assessed species being in decline to now 41% at risk of extinction, making them the most threatened vertebrate class (IUCN, 2024; Wake & Koo, 2018). Although stated in a recent publication by the IUCN that the total amphibian extinction is now at 37 species (Re:wild et al., 2023), others suggest as many as 200 frog species have been lost (Alroy, 2015). The aforementioned high rate of species discovery has led to an inability to keep our knowledge of amphibians up to date. Species are not being described, classified, and given an ICUN rating with the limited funding dedicated to frog conservation and the limited taxonomists currently available (Tapley et al., 2018). There are 61.3% of amphibians which are either not evaluated or have out of date assessments. In contrast, the conservation assessments in birds and mammals are completed or up to date in 99-100% of species (Tapley et al., 2018). Countries who have the most amphibian diversity are also the countries that have the highest number of new species discoveries, as well as the highest proportion of out of date descriptions (Tapley et al., 2018). Of the top 10 most amphibian-rich countries with the highest rate of new species discovery, only

Madagascar is up to date in its conservation assessments. It is estimated that between US \$884,709–\$1,656,980 is required to bring the amphibian conservation status assessments up to date (Tapley et al., 2018).

Global development has caused an increase in anthropogenic activity, which has led to environmental changes that negatively affect amphibians due to their unique morphological, behavioural and reproductive traits (Catenazzi, 2015; Tapley et al., 2018; Wells, 2007). Firstly, they have a smaller geographic range than other vertebrates (Grenyer et al., 2006; Nowakowski et al., 2017). The natterjack toad (*Bufo calamita*) experienced severe habitat loss from the urban and agricultural development of its native heathland in the United Kingdom (Wells, 2007). Deforestation and habitat loss also lead to habitat fragmentation, reducing population sizes. Small populations are more fundamentally affected by genetic issues (such as genetic drift, a loss in heterogeneity, inbreeding depression) and demographic and environmental stochastic events which may result in extinction vortices (Catenazzi, 2015; Gilpin & Soulé, 1986). Their permeable skin can serve as an avenue for the absorption of toxins and pollutants from the environment (Llewelyn et al., 2019). Due to amphibians' dependence on water, changes in temperature and increased prevalence of droughts also have an effect (Wells, 2007). Additionally, globalisation has facilitated the spread of the chytrid fungus (*Batrachochytrium dendrobatidis*), which is decimating frog species across the globe (Catenazzi, 2015; Wake & Koo, 2018). Finally, amphibians can be disproportionately affected based on their life histories. For anurans, those most affected are larger in size with smaller reproductive outputs, who are typically specialist species (Nowakowski et al., 2017). If populations decline, generalists are likely to move into these vacant niches, and may not serve the specific ecosystem functions previously provided, affecting the ecosystem's productivity (Nowakowski et al., 2017).

Previous work

In 2010, a previous student at Glasgow University, Michael John Elliott, explored the trends in global frog discovery between the 2001 and 2009. He proposed potential future exploration, namely, to expand the time scale of the study to see if the rate of frog discovery tapers off, indicating that the majority of species have been found. Another suggestion was to analyse how traits of newly discovered frogs differ from previously discovered frogs (Elliott, 2010). This would show if recently discovered species tended to be smaller, more cryptic, or fossorial, requiring increased research effort to find. This project, alongside exploring the state of taxonomy related to amphibians, also aims to expand on Elliott's previous research, by comparing amphibian discovery trends from 2000-2009 (henceforth referred to as decade 1), to 2010-2019 (decade 2), and investigating any changes in the physiological traits exhibited by frogs between decades.

As mentioned previously, conservation and taxonomy are linked; it is not possible to protect a species that has not been identified or named, and therefore taxonomy's success plays a role in the success of amphibian conservation (Mace et al., 2004; Nicolson et al., 2023; Tapley et al., 2018). It is therefore hoped that in this investigation, by using a large dataset of newly described species, the characteristics of amphibian taxonomy and the factors affecting it, will be apparent.

Aims

1. To determine the top countries where amphibian discovery is occurring and to determine how many amphibian species have been described between 2000-2019 and if this changes between decades.

- 2. To determine the proportion of amphibian species discovery taking place within protected areas, and the possible factors affecting this.
- 3. To determine the presence of trends in frog species' traits in correlation to their decade of discovery.
- 4. To determine the top countries responsible for carrying out new amphibian discoveries, the possible factors affecting this, and the top countries responsible for providing funding to enable amphibian discovery.
- 5. To determine the proportion of research published on newly discovered species that is open access between 2000-2019, and if this changes between decades.
- 6. To determine the proportion of discovered amphibian species with genetic sequences available on GenBank between 2000-2019, and if this changes between decades.

Methods

This study collated information from many research papers, as a meta-analysis. Although there is a trade-off in the loss of detail from individual papers, a meta-analysis is beneficial as it increases the breadth of results, reduces potential biases and resolves any uncertainties that may be present in individual papers.

Data sourcing and processing

Most of the data was obtained from the ASW online database, curated by Darrell Frost, linking information on amphibian species to primary literature (Frost, 2024). The ASW database is not available to download as structured data, so individual web pages were

downloaded, and data extracted by my supervisor, Professor Roderic Page. As part of that process, citations of individual papers were matched to the CrossRef (1999) database to retrieve DOIs (Digital Object Identifiers) for those papers. Metadata from each article, as well as author ORCIDs (Open Researcher and Contributor ID), and funder DOIs was retrieved using the DOI resolver (DOI Foundation, 2024). Using this metadata, the DOIs could be run through specific APIs, such as Unpaywall (Orr et al., 2024), to determine if there was an open access version of the paper available online, and Geonames (Wick, 2015), to link a two letter country code identifier to the location data from ASW. From these country codes, the corresponding continent was manually added. As South America holds 5-12% of the world's biodiversity (Lipińska et al., 2024), it was allocated its own continent to examine trends more closely. Funder DOIs were checked against Wikidata to where the funding body is based (Wikimedia, 2012). The availability of genetic data in GenBank was gathered by querying if the DOI was present in a sequence entry (NIH, 2024). Researcher nationalities were initially derived from the inputted affiliation of an author to their ORCID. This proved challenging as many either did not have an ORCID, or had a so-called 'ghost ORCID' with only minimal information present (Teixeira da Silva, 2021). To increase the volume of this data, my supervisor instead used affiliation data directly from the papers to find where the author was based; using the Research Organisation Registry (ROR) to parse affiliation strings and match them to a specific ROR ID (ROR, 2016). Those that were not matched were then run through the CERMINE API to extract country codes (Tkaczyk, 2024).

Once all the cleaned ASW data was obtained from Professor Roderic Page, it was processed using DB Browser for SQ Lite (Piacentini, 2021). SQLite is a large database software appropriate for handling the large amount of data (Hipp, 2023). Based on the aims outlined previously, the data was collated, then joined or filtered into different formats.

Alongside the data directly from ASW, trait data was obtained from the Amphibian Traits Database (Huang et al., 2023), a database of physical characteristics, particularly measurements of amphibians. It was hoped that the AmphiBIO database from Oliveira et al. (2017) could also be used due to the range of physiological and behavioural traits included in the database but there was insufficient information available for species described after 2011. The species names, and relevant trait (which was selected to be snout-vent length (SVL) measurements) was filtered from the Amphibian Traits Database. The investigation of trait data was only carried out for frogs and toads (Anura). This decision was made due to their comparable body plans, allowing more straightforward comparison (Wake & Koo, 2018). Lastly, protected area data was obtained from the World Database on Protected Areas (IUCN & UNEP-WCMC, 2024). It was imported to QGIS (2024), along with the GPS coordinates parsed from the ASW data. The number species discovery coordinates found within protected area vectors were counted, isolated and compared to points outside of the protected area vectors.

In total there were 25,240 papers describing amphibian species scraped from ASW. From these, 7,690 were extracted for the relevant timeframe of this study, 2000-2019. There was metadata available from the DOIs for 5,446 papers, and location (country) data available for 1,371 species, while location (latitude/longitude) data was available for 1,315 species. To get precise results for the specific number of new amphibian descriptions per year, the exact publishing date of new species description was used to filter the data further, and it was found that there were 2276 papers describing new species between 2000-2019. There were 1473 different SVL measurements taken from the Amphibian Traits Database, for 902 different species.

Data analysis

The data was all analysed using R (v4.3.1) (RCoreTeam, 2021; RStudioTeam, 2021). General(ised) Linear Models (GLMs) or Generalised Linear Mixed Models (GLMMs) were used for all statistical tests. Data was modified using dplyr (Wickham et al., 2023). For all GLMs, the real data was compared to simulated data from the model using functions within DHARMa (Hartig, 2022). If data was count data, and showed over-dispersion, the data would be modelled as negative-binomial, from the MASS package (Venables & Ripley, 2002), rather than Poisson. Additional detail, including algebraic structures of the complex models, is given below:

i. New amphibian descriptions between decades

A GLM was carried out to see if the number of papers published describing new species per year were significantly different in decade 1 and decade 2, suggesting a change in the number of amphibians being discovered. A negative-binomial distribution was used due to over-dispersion. The response variable was a count of the number of published papers per year, and the explanatory variable was decade, which was categorical, with 2 levels (decade 1 and 2).

ii. Location of new amphibian discoveries between decades

$$
\log(f_i) = c + \alpha_j + \beta_k + \gamma_{jk}
$$

 $i = 1 ... 108$ $i =$ Africa...South America $k = d1, d2$

A GLM was carried out to investigate the number of new amphibians described per continent between decades. The response variable was count data, of the number of species described per continent per year, and was over-dispersed, so a negative-binomial distribution was used. The explanatory variables were continent, α , (categorical: Africa, Asia, Central America, Europe, North America, Oceania, and South America), year, β , (categorical: decade 1 or 2), as well as interactions between continent and year, γ . Interactions were tested to see if the numbers of species being found per continent changed between decades. A post-hoc test, a pairwise comparison using the emmeans package (Lenth, 2024), was run to compare the continents and decades to each other.

iii. Variables affecting if a species was found in a protected area

$$
\log\left(\frac{p_i}{1-p_i}\right) = c + \alpha_j + B_k + m_1 x_{1,i} + m_2 x_{2,i} + m_3 x_{3,i}
$$

 $i = 1 ... 1344$ $j = d1, d2$ $k = B_1 ... B_{72}$

This GLMM, created using the lme4 package (Bates et al., 2015) explored the variables affecting whether a species was found in or out of a protected area. The response variable was binomial, being a species either found in (1), or out (0) of a protected area. Country, B , was a random effect, with the fixed effects being decade, α, (categorical: decade 1 or decade 2), GDP, x_1 , (continuous), total area, km², of the country a species was found in, x_2 , (continuous), and proportion of protected area of the country, x_3 , (continuous). The variables were tested for collinearity using Variance Inflation Factors (VIFs) from the car package (Fox & Weisberg, 2019), all of which were less than 1.2. Gross Domestic Product per capita (GDP), in \$USD, was obtained from the WDI package (Arel-Bundock, 2022). Total country area was obtained from Worldometer (2024), and the proportion of protected areas were obtained from The World Bank (2024). Due to the different scales of the variable values, the log of GDP, and the square root of total area were taken, and they were centred

and standardised. A Likelihood Ratio Test (LRT) was not carried out as all variables were deemed important to investigate.

iv. Effect of decade on body size

The response variable for this GLM was snout-vent length (SVL), recorded in millimetres, which was log transformed, and was distributed normally. Sex (Female or Male) and decade (1 or 2) were explanatory variables and were both categorical.

v. Effect of GDP and researcher number on reach of that country's research

$$
\log(f_i) = c + m_1 x_{1,i} + m_2 m_{2,i} + m_3 m_{3,i}
$$
\n $i = 1 ... 33$

For this GLM, the response variable was the research reach, measured as a count of the number of unique destination countries visited by researchers from an origin country. The explanatory variables were continuous, with the number of total researchers recorded from the origin country, x_1 , the GDP of the origin country, x_2 , (Arel-Bundock, 2022), and species richness, x_3 . Species richness was calculated as the total number of species observed in all destination countries visited by a country's researchers between 2000-2019, derived from the ASW database. The variables were checked for collinearity with all VIFs being less than 4.75. An LRT was carried out, using the lmtest package (Zeileis & Hothorn, 2002) to compare the amount of variation explained between different nested models.

vi. Effect of decade on a paper being open access

This GLM was run to see if there is a significant difference between the proportion of papers that were open access, and decade. The model was binomial, with the response variable

being a paper that was either open (1), or not open access (0). The explanatory variable was decade, which was categorical with 2 levels (1 or 2).

Graphs were made in RStudio using ggplot2 (Wickham, 2016), or for Fig. 8, using the website SankeyMATIC (Bogart, 2024). Maps additionally were created using Geocharts in Google Sheets (Google, 2024) or were screengrabs from QGIS (2024), using map data from Natural Earth (2009).

Results

New amphibian descriptions between 2000-2019

Between 2000-2019, there were 2276 new species described, or 114 per year. There were lower rates of description, less than 100 descriptions per year, until 2005 (Fig. 2a). For 2005- 2019, the rate of species descriptions over time appears to remain fairly constant (Fig. 2b). There was a larger range in the number of species discoveries per year in decade 1, compared to decade 2 (Fig. 2c.). When comparing between decades, it was found that there was a significant increase between decade 1 and decade 2 ($z_{1, 19}$ = 2.07, P=0.0385).

Figure 2: Number of new amphibian species descriptions over time. (a) a line graph showing the number of species described per year between 2000-2019, (b) a line graph showing the cumulative increase in species described over time between 2000-2019, (c) a boxplot showing the number of species described per year per decade.

Locations of amphibian discovery

From the data available, 1371 amphibians were discovered in 77 countries between 2000- 2019 (Fig. 3). Species were found on all continents except for Antarctica. The countries with the most amphibian species discoveries are Brazil, Papua New Guinea, India, China, and Madagascar. Between decades, the top countries of discovery were found to remain similar. In some countries, there does appear to be a substantial change in species discovery over time, such as in Sri Lanka, where 37 discoveries were observed in decade 1, but only 4 in decade 2.

Figure 3: Global distribution of new species discovery. The location of amphibian discoveries, that were later described and published between 2000-2019. The darker the colour orange, the more species discovered in that country. There was a maximum of 230 species, and a minimum of 1. Countries in light grey had no data.

Looking at the location data more broadly, the continents leading amphibian discovery are Asia, with an average of 31.8 descriptions per year followed by South America (20.2 descriptions year⁻¹), and Africa (9.65 descriptions year⁻¹) (Fig. 4a). The rate of species descriptions tapers off slightly in South America towards the end of decade 2, conversely it accelerated in Asia in 2004 and 2009, and in 2010 for Africa and Central America (Fig. 4b). Additionally, North America had higher levels of species description early in decade 1. There was not a significant change in the number of new amphibian descriptions between decades $(z_{13.94}=1.69, P=0.0902)$. From the pairwise comparison, it was also found that there was no significant increase or decrease in the number of species found per continent between decade 1 and 2 (Appendix A). When looking at the three continents with the most amphibian descriptions, it was found that there was no significant difference between Asia & South America in decade 1 (Tukey: z-ratio=1.95, P=0.796) or 2 (Tukey: z-ratio=3.14, P=0.0952), there was a significant difference between Asia & Africa in both decade 1 (Tukey: z-ratio=6.27, P<0.0001), and 2 (Tukey: z-ratio=6.29, P<0.0001), and there was a significant difference between South America & Africa in decade 1 (Tukey: z-ratio=4.43, P<0.001) but not decade 2 (Tukey: z-ratio=3.25, P=0.0691). Central America, Europe, North America and Oceania all had relatively low numbers of new amphibian discovery per year, and it was found that they were not significantly different from each other in either decade. All interactions tested of each continent between decades, and comparisons of continents at each decade can be found in Appendix A.

Within the countries of amphibian discoveries, there were a subset found in protected areas. For decade 1, 40.30% of species (n=593) were found in protected areas. This number decreases to 36.93% in decade 2 (n=723) (Fig. 5). Many of the discovered species that were outside of protected areas were often still found near protected areas (Fig 6). It was found that neither decade $(z_{6,1337}=-0.0720, P=0.942)$ nor a country's GDP $(z_{6.1337}=0.0380, P=0.970)$ had any significant effect on whether species were found inside a protected area. Although it appeared that the total area of a country had a negative effect on species being found in protected areas, this was not significant $(z_{6,1337}=-1.69, P=0.0906)$. It was found, however, that the proportion of protected areas in a country positively affected if a species was found inside one $(z_{6,1337}=3.374, P<0.001)$.

Figure 4: Number of new amphibian species descriptions per continent over time. (a) a line graph showing the number of species described per continent year between 2000-2019, (b) a line graph showing the cumulative increase in species data describing new species between 2000-2019. (c) a boxplot showing the number of species described each year on each continent per decade.

Figure 5: Location of new amphibian species descriptions in relation to protected areas. Map showing the distribution of species discovered within (blue) and outside (orange) protected areas between (a) 2000-2009 and (b) 2010-2019.

Figure 6. A close up of distribution of species in and outside protected areas. Map showing the distribution of species discovered within (in blue) and outside (in orange) of protected areas (light green) between 2000- 2019. This map shows a selected subset of protected areas, in Ecuador, at a scale of 1:4100000.

Traits exhibited in discovered anurans

SVL was found to be significantly different between sex, with males being smaller ($t_{(2, 1415)}$ =-6.78, P<0.001). SVL also significantly decreased from decade 1 to decade 2 ($t_{(2, 1415)}$ =-3.55,

Figure 7: Box plot showing body size (SVL) of new Anuran species described in decade 1 and decade 2. The SVL data was log transformed and split into female (orange) and male (blue) sex data. Juvenile data was excluded.

Location of authors publishing amphibian descriptions

Researchers were from 33 different countries (Fig. 8), with most coming from the United States and Brazil. The US showed the widest spread of research destinations, with a total of 16 separate countries receiving researchers. In contrast, there is a high percentage, 90.6%, of Brazilian researchers carrying out research in Brazil. This trend can also be seen in China, Ecuador, Mexico, and Indonesia, where most researchers carry out research in the country where they live. For other countries, this is not the case, as seen in Nigeria, Mozambique, and Madagascar, where most researchers come from India, South Africa, and Germany,

respectively. Upon carrying out an LRT for the different nested models, a simpler model, including only number of researchers and GDP, was found to have no significant difference in explaining the variation in the data to the more complex model, so the complex model was disregarded ($χ=3.46$, P=0.0630). The number of researchers ($z_{2,30}$ =6.580, P<0.001), and the GDP of the origin country ($z_{2,30}$ =2.378 P=0.0174) both had significant positive effects on the number of countries visited by researchers.

Figure 8: Sankey diagram showing the origin of researchers (left), and the location where they carry out amphibian research (right). The numbers on the left correspond to the number of authors listed on a published paper published with affiliations from those origin countries between 2000-2019 and the number on the right is how many authors are credited for working on a paper about a species found in that country between 2000-2019.

Location of funders for amphibian research

There was limited data available for the funders, with 149 funders from 13 countries being

traceable through their DOIs (Fig. 9). Despite this, it was found that the United States,

Denmark, Germany, and India were the main countries involved in the funding of amphibian research.

Figure 9. Global distribution of funders of research on amphibian discovery between 2000-2019. There was a maximum of 39 funders found within one country, and a minimum of 1. The darker the yellow, the more funding bodies for amphibian discovery there are within that country. Countries in light grey had no available data on funders for amphibian research between 2000-2019.

Open access

Of the 3568 papers with data on access status available (Fig. 10a), it was found that 1497, or 41.96% were open access. In the breakdown of countries, Bulgaria, Cyprus, Greece, Guinea, and Turkey all had a proportion of 1, showing that all their papers published were open access (Fig. 10b). They each had between 1-3 papers published. When broken down into decades, it was found that in decade 1 (n=1456), 37.71% of published papers with data available were open access, compared to 44.89% in decade 2 (n=2112). This was significant, with more open access papers published in decade 2 than $1 (z_{1,3566} = 4.267, P < 0.001)$.

Figure 10. Open access papers published on new amphibian species per country between 2000-2019. (a) The total number of papers published describing new amphibians by an author in their affiliated country. The darker the green, the higher the number of papers published from authors within that country, (b) The proportion of open access papers published on new amphibian species by an author in their affiliated country. The darker the green, the higher the proportion of open access papers from authors within that country.

GenBank

Of the 5446 papers analysed, 705, or 12.9% had associated GenBank sequences linked to their DOIs. For these 705 papers, there was a total of 11,121 genetic sequences, with the number of sequences ranging from 1-20 sequences available per paper. The mean genetic sequences per paper was 15.77. There were 266 papers with associated sequences in decade 1, increasing by a factor of 1.64 in decade 2, with 437.

Discussion

It has been found that there is still a high number of new species being described per year, and that there was a significant increase in the number of species described from decade 1 to decade 2. This may be due to the increasing uptake and use of genetic techniques to identify new species, as well as the increase in surveying techniques in the field (Kohler et al., 2005). There has also recently been the introduction of environmental genetic surveying to find eDNA. eDNA is genetic material that has been shed into the environment from

organisms and can be detected in water, sediment, and air samples (Breton et al., 2022). This allows the detection of the presence of many new species, without the need to find them and collect DNA directly (Sun et al., 2024). This novel technique suggests that amphibian species discovery may increase further in the coming years. The increase of the availability of DNA of new species on GenBank that was observed in this investigation, increasing by over 1.5x from decade 1 to decade 2, supports the idea that there has been an increase in the use of genetic techniques for species identification between 2000-2019.

The countries observed to have the highest number of species discoveries is as described in recent literature. Brazil was the country with the highest number of new species descriptions in both decade 1 and decade 2, which matched the literature from Segalla et al. (2019). Madagascar and Papua New Guinea were noted as island nations with high amphibian abundance (Wake & Koo, 2018), and they showed high levels of amphibian description in this study. Findings such as the decrease in species discovered over time in Sri Lanka could be explained by a change in research effort over time, or simply that most amphibians have now been discovered. To validate this, there should be further analysis of whether other factors have also changed between decades, such as the number of herpetologists completing research in Sri Lanka. Additionally, Costello et al. (2013) found that Asia and South America had the highest level of new species descriptions, which also is in line with the findings of this investigation. Despite Asia having a greater overall number of species descriptions, it was found that there was no significant difference in either decade between Asia and South America in species descriptions per year. This is an interesting finding in regard to the well-established fact that South America is the continent with the highest amphibian species-richness (Vasconcelos et al., 2019), and it would be expected that South America had significantly higher levels of amphibian description than all other

continents. It would therefore be interesting to investigate if there is the possibility of Asia overtaking as the most species-rich continent, if the difference between species discovery in Asia and South America were to increase further. Both Asia and South America having a significantly higher number of amphibian descriptions than Africa in decade 1, and Asia in decade 2, further confirms that Asia and South America are the leading continents in amphibian discovery. It could be suggested, however, based on the results that amphibian description of African species was increasing in decade 2, that it could be increasing in the number of amphibians described over time, but further study into the coming decades would be needed to confirm this. It has been found that approximately 114 amphibian species were described per year, with the top countries as Brazil, Papua New Guinea, India, China, Madagascar, Vietnam, and Sri Lanka.

Most new species were discovered outside of protected areas, with the number of species found inside protected areas decreasing from decade 1 to decade 2. This low amount of in-protected area species discovery could be due to several factors. One such reason could be that the protected areas were declared as protected due to the high level of biodiversity found there previously, and thus, there are not many new species left to still be discovered. An alternative reason is that it may be challenging to obtain permits to carry out research, sample species, and even enter these areas, due to their increased importance for a nation's biodiversity. Upon closer observation of the protected area and species discovery-location data (Fig. 6), it was found that many species deemed outside protected areas were actually found along the margins of or just outside these protected areas. Brattstrom (pers. com, 2024) has proposed this is the result of researchers avoiding the administrative burden of gaining permits to carry out research in protected areas. The number of species found near these protected areas shows the high biodiversity and

importance of these areas, whether they are protected by governments or not, begging the question of whether protected areas should be moved or expanded in order to encompass as much biodiversity found in these regions as possible. The findings of the GLMM showed that the proportion of protected area in a country had a significant positive effect on whether species were found in that area. This can be attributed to the fact that a greater proportion of protected area encompasses more land, which in turn supports more species, and so the likelihood of these species being undiscovered is also higher.

As the broader behavioural and physiological data from AmphiBIO was excluded due to lack of coverage, only SVL was compared between decades as a trait. SVL was found to get significantly smaller from decade 1 to decade 2. This may suggest that as time passes, and more frog discovery takes place, the remaining undiscovered frogs tend to be more inconspicuous, meaning they are discovered later when research effort has had to increase. This is a trend that has been observed previously (Brackley, 2022).

Species discoveries were carried out by researchers of many different nationalities. The research reach by countries was found to increase with GDP. Researchers from wealthier countries, such as the United States, tended to travel to many different countries and it can be observed that these destination countries tended to themselves be lower income, such as the DRC, Ethiopia, and Guyana. In contrast, middle income countries, who likely had fewer resources to travel the same distances as high-income countries, tended to complete research in their own countries, as seen in Brazil, Ecuador, and Thailand. Those in lower income countries did not complete much, if any research on amphibians either within or outside of their home country. These findings are likely confounded by other factors such as level of development of destination countries, and differences in amphibian species richness between the origin and host country, despite species richness not being excluded

from the model due to insignificance in the LRT. There is arguably a relationship between countries with more amphibian richness and their wealth. These relationships have been driven by the geographical location of that country (typically along the equator) and the historical interactions of their country with the West, such as through colonisation. This stunted the ability for these countries to be autonomous and build wealth to a level where they may begin to research the biodiversity within their borders (Collen et al., 2008; Oto-Peralías & Romero-Ávila, 2016; Thong, 2016). Additionally, the geographical position of the researcher's country of origin and its relative amphibian diversity likely also act as drivers for deciding the destination of research (Martin et al., 2012). For example, there is a lower amphibian species richness in Germany, and an abundance of trained taxonomists and zoologists, due to the higher rates of university education, often associated with a higher GDP (Collen et al., 2008; Meo et al., 2013). These two reasons could explain why German researchers would travel to a country like Madagascar to carry out amphibian research. In comparison, Brazilian researchers, who have much amphibian richness within their county's own borders, would feel less need to travel internationally to complete amphibian research, regardless of their country's wealth. Regarding the main countries who fund amphibian research, although there was limited information available, the trends were as expected. It was found that countries either with a higher GDP, such as the US, Denmark and Germany, or a higher amphibian species richness, such as Brazil and India, tended to be the countries providing more funding towards amphibian discovery.

The availability of research in the field of Zoology, as mentioned previously, is important for dissemination of knowledge to the public. This is especially true considering the high level of public interest in the environment (Manfredo, 1989; Mittermeier et al., 2021). Additionally, the interest within the public of the environment will likely only

continue to increase as the effects of climate change worsen. It was found that almost 42% of papers describing new amphibians were open access, and therefore available to the public. Although the proportion of open access papers increased from decade 1 to decade 2, it can be said that effort should be made moving forwards to increase this proportion further, to ensure information about nature is widely available, accurate and up to date (Goodschild van Hilten, 2022).

The final point of investigation in this study was the proportion of newly described species that had genetic sequence data publicly available. It was found that only a small percentage of analysed papers had associated genetic sequence data in GenBank. This investigation only considers if the GenBank entry had an associated DOI of the relevant paper linked to it, and not if the authors or title of the paper was credited alone, so it is likely the actual number of amphibian sequences of newly described papers available in GenBank is higher than was found in this investigation. The inclusion of sequence data into GenBank can improve the understanding of relationships between species, which in turn can inform suitable conservation efforts.

Limitations

There are some flaws within the methodology and collected data. The primary reason for this is because all data sourced for this investigation was collected by myself and my supervisor, Professor Roderic Page, and due to limitations with time, we were not able to ensure the data was clean and without errors. There are unfortunately a few steps where errors could arise throughout the data scraping, and collection. Errors were for example encountered when initially mapping the species discovery coordinates into QGIS, and many points were found to be in the incorrect locations, such as in oceans, and in deserts, both

unlikely locations for high frog abundance. This was found to have been caused by the code to withdraw the GPS co-ordinates from the papers interpreting them in the wrong format. Efforts were made to remove these incorrect points, but it is possible mistakes slipped through both with location data, and other data less straightforward to visualise. These mistakes were also difficult to pick up due to the large volume of data being processed. In addition to the mistakes in the location data, there were also limitations in its availability. Firstly, if the location was a written description, rather than GPS coordinates, then it was not recognised by the scraping code. Secondly, not every paper analysed had included the location of the amphibian discovery. This could be due to a variety of factors, for example, simply not deeming it necessary to include, but it also may be excluded to avoid sharing the locations of unique and rare amphibians. Sharing locations of amphibians who are being overharvested, such as the Lao newt (*Laotriton laoensis*), could lead to their further capture and sale as part of the illegal wildlife trade (Phimmachak et al., 2012). There are also limitations within the findings of the analysis of SVL trait, as the level of relatedness between species was not considered and the results may be phylogenetically biased. It may have been the case that more closely related species, who are more similar in morphology and behaviour were discovered at the same time due to other factors (such as habitat type or colouration), and their SVL cannot be tested without taking these factors into account. With more time, updating both AmphiBIO and the Amphibian Traits Database in the future would be useful, to increase the options for trait analyses. Additionally, it is likely more locations could be pulled directly from papers, and closer analysis on the availability of sequence data of new species could be carried out, rather than relying just on the DOI being linked to the sequence entry.

Conclusions and future work

This investigation has found that there are still high rates of amphibian discovery, and undoubtedly many species still in the wild awaiting discovery and description. Amphibians are an important group for ecosystem function and as bio-indicators of ecosystem health (do Amaral et al., 2019). It is therefore important that there is the taxonomic framework in place for these species' features, their interspecific relationships, and their behaviour to be researched. There is a necessity for amphibians to be assigned a conservation status, especially considering the drastic ongoing environmental changes, and the impending pressure on species worldwide (Tapley et al., 2018). Despite the importance of species discovery for scientific advancement, there are also challenges in completing research on biodiversity. Research inherently is about discovering new and untouched places, to find what has previously been unknown. This comes with challenges, as infringement of wild places tends to lead to their development, which in turn may negatively affect the flora and fauna living there (Clémençon, 2021; Coria & Calfucura, 2012; Zeppel, 2006). Furthermore, much is still unknown in less developed countries, due to insufficient funding, lack of infrastructure, and political upheaval (Collen et al., 2008). Additionally, and because of this, existing research is biased to the West, where most researchers come from and have therefore thoroughly explored these more developed areas (Christie et al., 2021; Martin et al., 2012). Development of low-income countries is vital for improving welfare and livelihoods, and the possibility of economic advancement enables inhabitants to begin carrying out ecological research that has been previously neglected due to other, more pressing issues.

It does appear that there is movement in the right direction, as the findings from this investigation show an increase in open access papers, an increase in availability of genetic

sequences, and an increase in the number of descriptions of new amphibians over time. Despite this, there is still opportunity for further work. Generally, authors should be encouraged to fill in their ORCIDs, to reduce the number of ghost ORCIDs and increase validity and integrity of academic work (Teixeira da Silva, 2021). Additionally, the findings of investigations such as this one can provide opportunities to identify under-researched areas, by comparing regions that are undergoing high levels of exploration and comparing them to known regions of high species-richness. This could highlight geographical pockets that are understudied and can then be targeted to find more species. Additionally, the use of drones for conservation has been suggested to overcome some of the geographical and technical challenges of conservation management (Sandbrook, 2015). There could be further development of this technology for biodiversity research, to collect data on new species, for example by collecting eDNA without the need to infringe on wild places or overcome the technical difficulties of carrying out field research.

Finally, an additional challenge for amphibian discovery is the sheer number of amphibian species, meaning difficult decisions need to be made regarding the allocation of research effort. Specific groups deemed to be less taxonomically important to the functioning of an ecosystem or maintaining biodiversity will need to be sacrificed (Christie et al., 2021; Drew, 2011). These decisions will need to be collectively made by the entire taxonomic community. Taxonomic expertise is required to ensure quick decisions can be made while remaining well-informed. In the 1990s, a species of spider mite, responsible for infesting farmed crops, was misidentified in the south of Africa and while costly plans were being developed to control it, it was identified by taxonomists to be an invasive mite from South America, and was able to be quickly, and cheaply dealt with using an imported predator (Drew, 2011). This is just one example of how having educated taxonomists can

help deal with widespread problems affecting various aspects of society, and illustrates how support for taxonomy is vital for the success of conservation (Drew, 2011).

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Appendices

Appendix A

The results of the Pairwise Comparison of the interactions of continent and decade in number of amphibian descriptions per year (see model ii in *Methods*). The values below are for the continents being compared per decade (in **bold**), and the comparison of continents within the same decade.

