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## The hidden history of humming frogs

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# The hidden history of the humming frogs

In the depths of Brazil's Atlantic Forest, researchers are battling to identify species before they are lost for good. **Dr Rafael O de Sá**, Professor of Biology at the University of Richmond, Virginia, USA, specialises in a group known as the 'narrow-mouthed frogs' and has already found four species new to science.

**T**he amphibians (frogs, toads, newts, salamanders and caecilians) are one of the least-understood groups of vertebrates on earth, and one of the most vulnerable. In the Neotropics – a poorly-explored region extending from Mexico and the Caribbean to the southern tip of South America – urgent conservation efforts are required to protect amphibians and their rapidly disappearing habitats. However, the species cannot be protected unless we know that they exist, how to identify them, and where to find them.

Dr de Sá's research aims to fill the gaps in our knowledge surrounding one particularly enigmatic group of amphibians – the 'humming frogs,' technically known as *Chiasmocleis*. *Chiasmocleis* is the largest genus of the narrow-mouthed frog family found in the Neotropics. The family Microhylidae has a worldwide distribution,

except Europe and Antarctica, and currently consists of over 600 known species.

Like all Neotropical narrow-mouthed frogs, *Chiasmocleis* live in burrows, emerging from their underground hideaways only during the breeding season. According to Dr de Sá, this is the best time to catch and study the elusive little creatures. He listens for the males breeding calls, follows the sound to its source and then catches the culprit.

## A FROG FAMILY TREE

The foundation of Dr de Sá's current research will be a 'family tree' – known as a 'phylogeny' – of all the frogs considered to belong in the *Chiasmocleis* group, built using genetic data. By examining the similarities and differences in the DNA of individual frogs from right across the range of the group, Dr de Sá can determine which are most closely related and which are more distantly related.

Genetic data provides a clearer understanding of relationships than phylogenies built using morphological characteristics, which provide conflicting evidence as similar features seem to have evolved repeatedly in unrelated species as a result of their similar burrowing lifestyles in this group of frogs. The final phylogeny will track the evolutionary history of *Chiasmocleis*, providing a framework to understand the evolution of humming frogs both genetically and morphologically.

Previously, Dr de Sá built a family tree for the narrow-mouthed frogs as a whole, to work out where within this extended family *Chiasmocleis* fits. Already, that tree has shown that not all species thought to belong to *Chiasmocleis* actually fit together in the same part of the family tree: three species posing as *Chiasmocleis* were really members of a nearby group called *Syncope*, and one further imposter

belongs to another distantly-related group, *Elachistocleis*.

Dr de Sá has officially renamed these species according to their correct affiliations, leaving him with a neat collection of 33 currently known species of *Chiasmocleis*, all descendants of a single common ancestor. He will now focus in on the relationships within this group, producing a revisionary up-to-date description of them all, known as a 'monograph'.

## CRYPTIC CREATURES

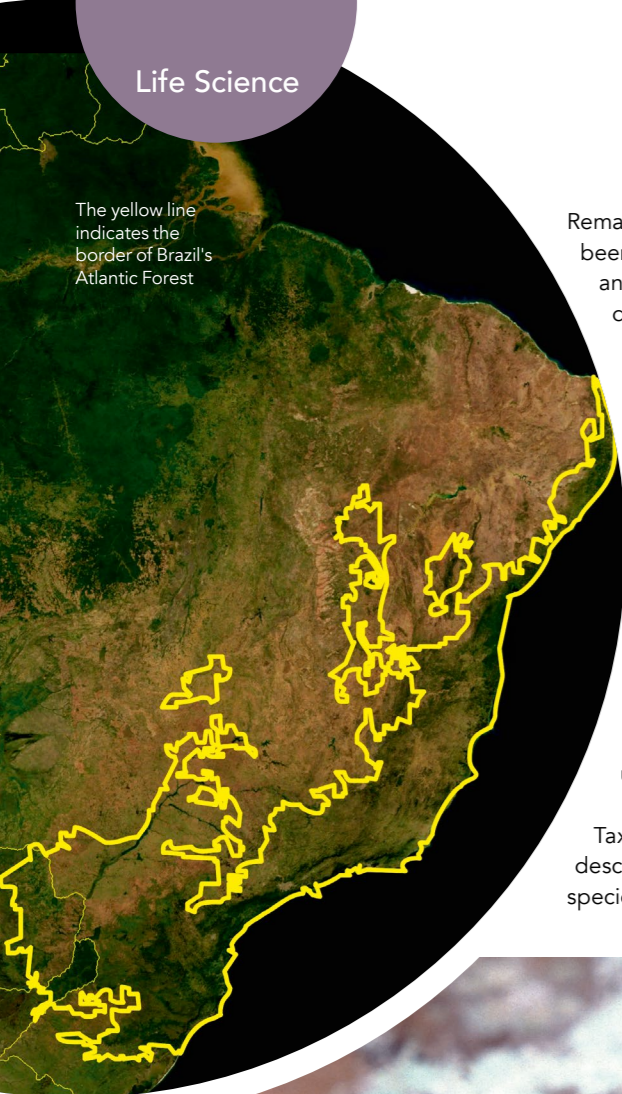
One of the first things Dr de Sá noticed while working on *Chiasmocleis* was that there were certain clusters of individuals with DNA similar to each other and distinctly different to the DNA of other groups. These clusters would suggest new species, yet they had not previously been identified as such based on their appearance. Dr de Sá calls these 'cryptic species.'



Species cannot be protected unless we know that they exist, how to identify them, and where to find them



The yellow line indicates the border of Brazil's Atlantic Forest



Remarkably, once the cryptic species had been identified from their DNA, a careful analysis of morphological characteristics did reveal some distinguishing features, which had been overlooked in previous studies. This included a small difference in size, foot webbing and skin pigmentation. As Dr de Sá says: "The molecular data guided us to reexamine ... the variation of external morphology ... and identified the characteristics that would allow us to recognise those populations as new species." This means that the new species can be recognised, not only in the lab, but in the field – a crucial factor for finding out more about them and ultimately saving them from extinction.

Taxonomists are the scientists that describe and name new species; this is how species become "known" and is key to all



**The new species can be recognised, not only in the lab, but in the field – a crucial factor for finding out more about them**

future biodiversity research. So far, Dr de Sá's team have formally described four new species of *Chiasmocleis* but Dr de Sá says his team have identified other potential new cryptic species, almost indistinguishable from one another to the naked eye. For Dr de Sá, this highlights the importance of using genetic data in biological studies: "The diversity [i.e., number of species] of Neotropical frogs goes beyond what we can identify through traditional morphological approaches." It is highly likely that the number of species of humming frogs will continue to grow.

#### A FOUNDATION FOR CONSERVATION

Dr de Sá's monograph will not only list the species of *Chiasmocleis* – cryptic or otherwise – it will also provide crucial information for their conservation. Firstly, a fully diagnostic description of each species and how it differs from the others, will enable future researchers to identify the frogs they come across.

Secondly, based on records of all the places the frogs have been found over the years, Dr de Sá will be able to estimate the range of each species. The humming frogs can be found throughout the lowlands of South America to the east of the Andes Mountains. However, like most animals and plants of the Neotropics, little is known about the area each individual species extends to cover. Combined with information on the extent of suitable habitat, this provides essential data for estimating the level of threat each species may face. At present, almost a third of species of humming frog are recorded as simply 'data deficient' on the IUCN's Red List of Threatened Species – because not enough is known about the species and their distribution.

Given the rate of habitat destruction in the Neotropics, Dr de Sá fears that even where frog species have been assigned a Red List status, this may be dangerously out of date. Many of the newly-described *Chiasmocleis* species are restricted to the Atlantic Forest of Brazil – an ecosystem under severe threat from habitat degradation and loss caused mainly by agriculture, such as cattle ranches and soybean plantations. Some estimates suggest that only 3.5% of the natural vegetation of the Atlantic Forest now remains. Dr de Sá's new monograph will provide invaluable information for researchers seeking to conserve these fascinating animals. Hopefully for the humming frogs, it will not come too late.

## Q&A

#### What is so special about working in the Neotropics?

Ecologically, i.e., variety of habitats, the Neotropics are extremely diverse. The three major groups of amphibians, (frogs and toads, caecilians, and salamanders) occur in the Neotropics. The region is home to 49% of the world's amphibian species and, of them, 39% are considered threatened, representing about 60% of the globally threatened amphibians. Consequently, it is critical that researchers and funding agencies allocate resources for research, training, and education focus to decipher the biodiversity of amphibians in the Neotropical realm and to understand the status of more than 120 species which are considered Critically Endangered and are possible extinct.

#### Why are amphibians so vulnerable to human activity?

Habitat destruction and loss is the major threat to all wildlife; recovery programmes can be effective if researchers have suitable habitat to reintroduce species in the wild. Amphibians have a 'naked' skin, a difference from other vertebrates with some protection such as scales, feathers, or hair. Furthermore, the skin of amphibians is a major site for respiration. Consequently, amphibians are particularly vulnerable to environmental pollution (e.g., pesticides, herbicides). Over the last 30 years, the spread of a pathogenic fungus has decimated amphibian populations. These threats compounded with human-caused climate change are driving the current amphibian mass extinction.

#### Can you describe how you first find a cryptic species from genetic data?

The analyses of genetic data from sampling of individuals across the distribution of a species provide a first approach to understand cryptic

speciation by the identification of 'clusters' of populations in a phylogenetic tree. Those clusters may represent new species. Subsequently, taxonomists explore other sources of data, e.g., detailed examination of external morphology, internal characters (e.g., bones, muscles), reproductive signals (e.g., calls, courtship patterns), developmental and life history traits (e.g., larval stages, egg lying sites), ecology (e.g., adult and larval environments, feeding), patterns of distribution and possible boundaries (e.g., rivers, valleys), to determine if those clusters represent new species.

#### How many more undiscovered species do you think are out there?

Undoubtedly each group of organisms has undiscovered species. Currently, *Chiasmocleis* consists of 33 known species; half of the known species have been described since 2003. Further analyses of *Chiasmocleis* will likely result in an increase of about 25-40% of the total number of species. Fieldwork is critical to document this disappearing biodiversity as well as to enhance our knowledge of the described species – information such as calls, larval stages, etc.

#### How does a monograph help in the conservation of species?

A monograph provides a summary of the information known for all species reported in a particular group (genus, family, order, etc.) of organisms. Then, this type of work provides a place where the non-taxonomist, students, and early researchers, as well as conservation efforts and policy decision making agencies can quickly refer to the available information for that particular group, including distribution, similar species, etc.

## Detail

#### RESEARCH OBJECTIVES

Dr de Sá's research focuses on the evolutionary biology of frogs. Within this, he utilises molecular and morphological approaches to phylogenetics, systematics, taxonomy, and development to understand the species diversity of frogs.

#### FUNDING

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#### COLLABORATORS

João F. R. Tonini

#### BIO

Humans, for the first time in our history, are witnessing the extinction of an entire group of animals: amphibians. Dr Rafael O. de Sá works with undergraduates at the University of Richmond and researchers and students around the world to document new species of frogs before they disappear. These scientists use interdisciplinary approaches to assess cryptic diversity in frogs.

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#### More info:

[news.mongabay.com/2017/03/three-new-frog-species-found-in-disappearing-atlantic-forest/](http://news.mongabay.com/2017/03/three-new-frog-species-found-in-disappearing-atlantic-forest/)

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**The Neotropics are home to 60% of globally threatened amphibians**

