Short-term population dynamics and microenvironment of *Andinobates aff tolimensis*

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Preliminary Report

Short-term population dynamics and microenvironment of *Andinobates aff tolimensis*

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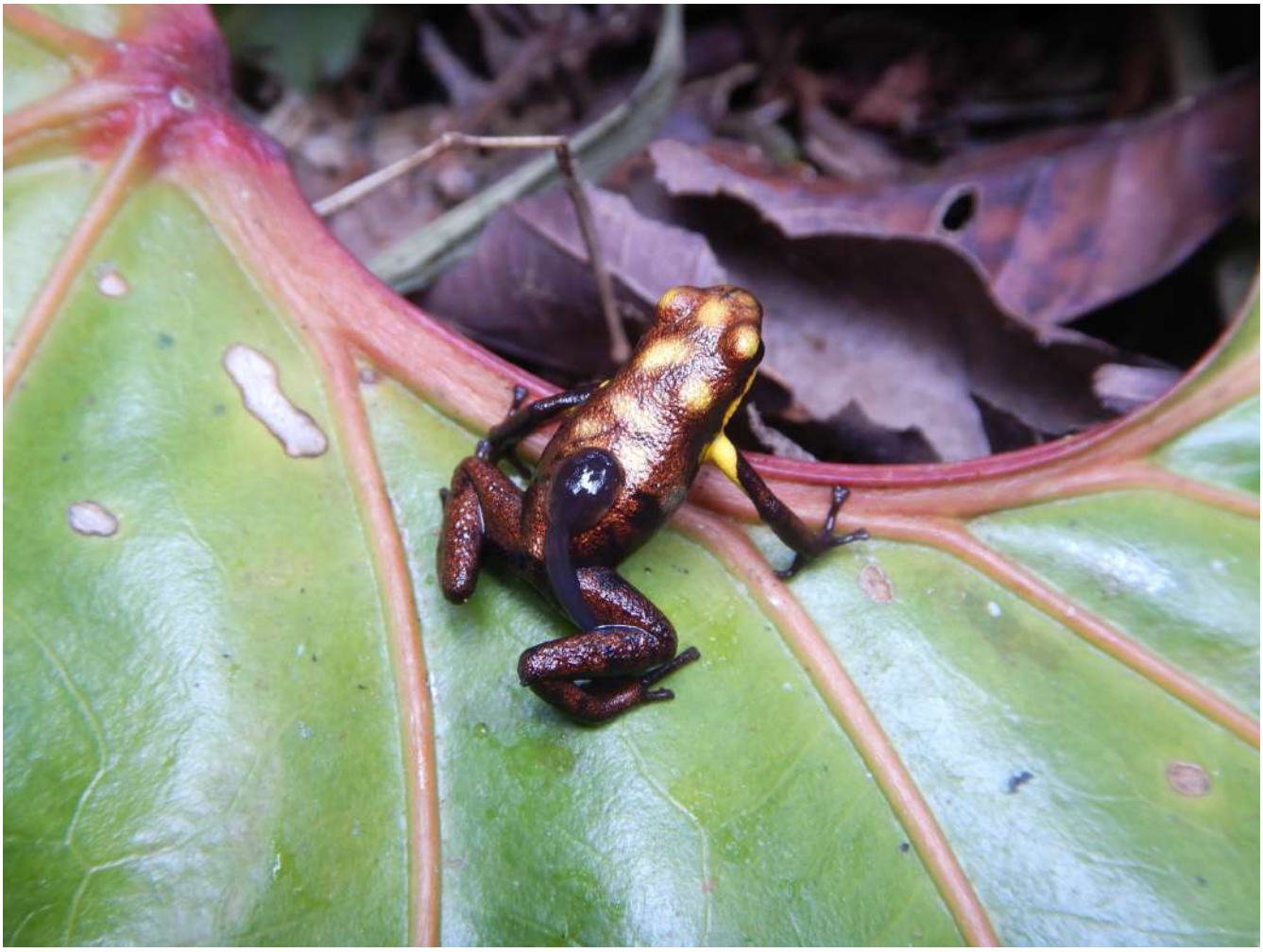
Summary

Our goal is to assess the population dynamics, behavior, and microenvironment of the threatened Andean poison frog *Andinobates aff tolimensis* in Padua village (Tolima, Colombia). This species has been only recorded in the locality of Frias in Falan (Tolima). However, our results suggest *A. aff tolimensis* has a small population in the Corregimiento (village) de Padua in Herveo (Tolima), around ten kilometers in a straight line from the type locality. The habitat in Padua is difficult to access despite it is only a little more than two kilometers from Padua. Although the habitat is protected, it is a really small forest patch. Our research was able to record microenvironmental and phenotypic data. Likewise, we have captured close to 400 records with 42 individuals recaptured at least one time. Nonetheless, the loss and fragmentation observed in the habitat deserve urgent measures. Currently, we have done seven field trips with good outcomes for the population dynamics analysis based on the extremely fragmented habitat. Likewise, we were able to record other threatened frogs and take microenvironmental data, tissues for DNA analysis, and phenotypical data.



Andinobates aff tolimensis carrying tadpole.

Introduction

Andean poison frogs of the *Andinobates* genus encompass sixteen species (Frost, 2023), fourteen of them occurring in the Andean Cordilleras of Colombia and lowlands of Colombian Choco. Unfortunately, most of them are threatened and there are deficient data for two species. *Andinobates tolimensis* is an endemic frog species from Central Cordillera in Colombia, specifically from Falan (Tolima, Colombia) (Bernal et al. 2007). This frog is categorized as Vulnerable by IUCN due to its range being highly restricted with all individuals occurring in a single forest fragment that does not exceed 0.5 km2 (IUCN, 2014). The species is diurnal and is restricted to inhabiting the ground between small burrows made by fallen logs and tree roots in a small primary forest. Although its original habitat is protected as a private reserve (IUCN, 2014), this species is pretty rare and nothing about its ecology, behavior, distribution, and evolution is known since the description. Likewise, *Andinobates* frogs have special interest due to most of them occurring on Tropical Andes in Colombia, the region with the greatest land cover change in Colombia (Amézquita et al. 2013; Amézquita et al. 2016).

This project focuses on the population dynamics of a new population of *Andinobates aff tolimensis* in the Corregimiento de Padua (Herveo, Tolima, Colombia). We have collected population information, morphology, calls, and microenvironmental temperature data. Furthermore, other amphibian species were recorded during the field trips, and several of them were threatened. Currently, we are collecting data to determine population dynamics and the spatial distribution for this probably new population of *A. aff tolimensis*. Likewise, we have enough tissue for genetic analysis and we hope in the next two months to determine the current taxonomic status of this population.

Our preliminary results increase data about the call behavior, spatial ecology, and natural history of little-known threatened frog species. This project highlights the importance of monitoring and conserving the forest in the Corregimiento de Padua (Herveo, Tolima), given the significant number of threatened amphibian species found during field trips.

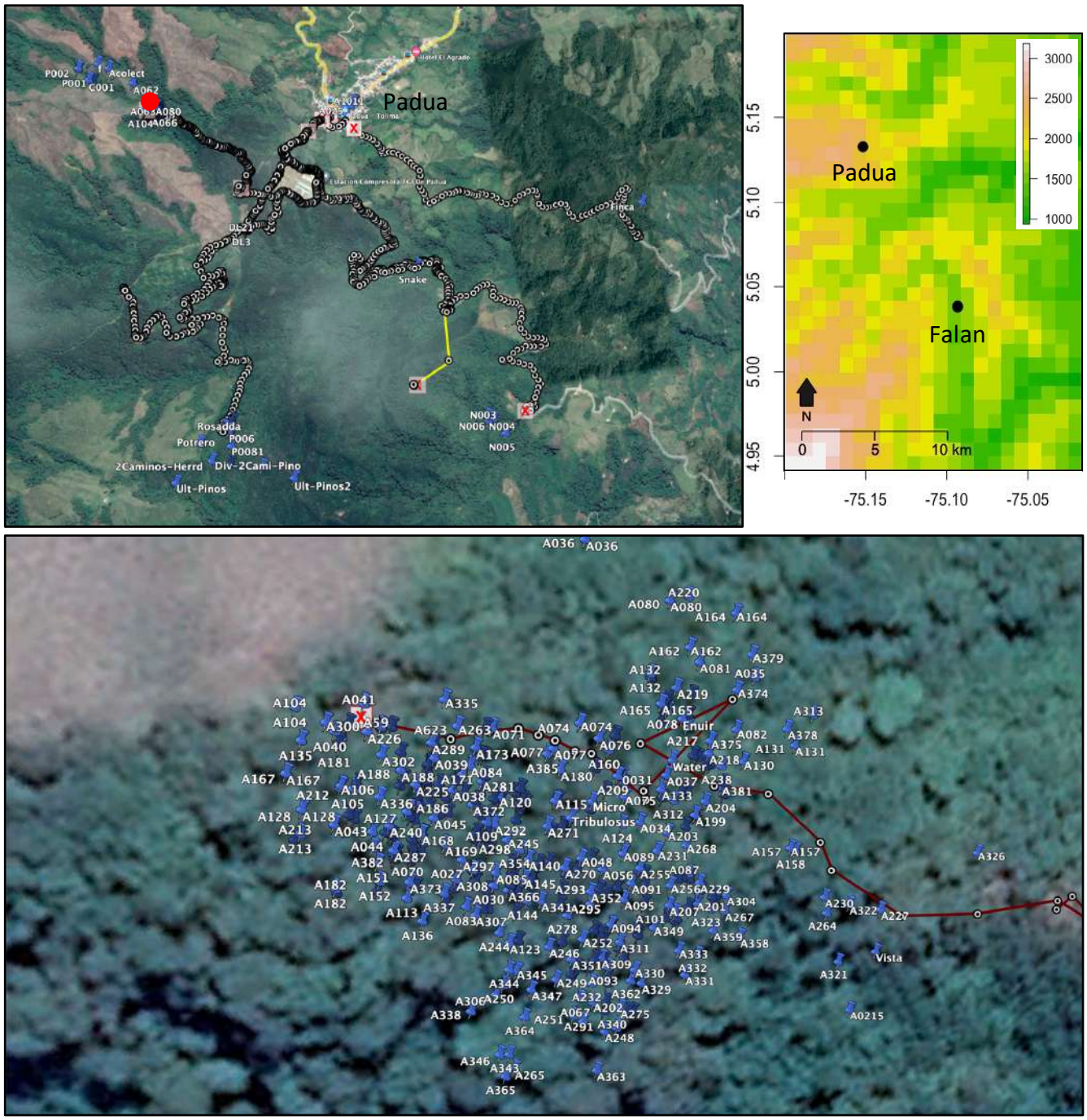


Figure 1. Area of study for population analysis of *Andinobates aff tolimensis* in Padua village (Tolima). Left, an Image from Google Earth shows the Padua village and the different paths to find *Andinobates populations*. The dark green land cover indicates Pinus patula crops. The red point is the area of the population analysis. This photo is outdated and much of what looks like a forest is actually pine crops that have already been felled and replanted with small pine shrubs. Right, the digital elevation model shows Falan, the type locality of *Andinobates tolimensis* and the probably new population in Padua. Bottom, the spatial distribution of capture-recaptures of *A. aff tolimensis* (red point).

Project members

Bibiana Tovar: Bibi has finished her undergraduate studies as a biologist at the Universidad del Tolima (Colombia) and currently works as an environmental consultant on different infrastructure projects. Bibi found new records of new threatened amphibians' species in Padua while working with the population dynamics of *Andinobates aff tolimensis*.

Zharik Sarmiento: Zharik is biology student from Universidad del Tolima and volunteer working with bioacoustics.

Maria Camila Murillo: Cami is biology student from Universidad del Tolima and volunteer working with bioacoustics.

Ricardo Medina: Ric is a Master in Evolutionary Biology from Universidad Nacional Autónoma de México (México) and a Biologist from Universidad del Tolima. Ric is working with spatial ecology, population dynamics, and found new threatened frogs, likewise mentoring bioacoustics to Zharik and Cami.

Aim and objectives

This project has the following seven objectives:

1. Determine short-term population dynamics of *Andinobates aff tolimensis* of a probably new population with capture-mark-recapture methods
2. Determine temporal and spectral features of the calls of *Andinobates aff tolimensis*
3. Determine the morphology of *Andinobates aff tolimensis*
4. Contrast new and published sequences of mitochondrial genes to determine the status of a probably new population of *Andinobates aff tolimensis*
5. Monitor microenvironment temperatures
6. Evaluate the presence of *Batrachochytrium dendrobatidis* (Bd)
7. Promote environmental education with students and different stakeholders, involving them in field trips and environmental workshops.

Although no objective has been changed there were partial adjustments during the implementation of the project.

Methodology

Field work sampling

Seven field trips were done between March and July 2023 in Padua (Tolima) with five days each for population analysis of the species *A. aff tolimensis* (**Fig. 1**, **Fig. 10**). Likewise, we record three new threatened frog species in the corregimiento de Padua. The first field trip was more extensive based on the fact that different sites were sought where to carry out the population study (**Fig. 1**). Although some calls of *A. aff tolimensis* were heard along different paths, only one point was found where there was a population that could be evaluated and that did not represent a risk. This population was located approximately 2.3 km away from Padua village with an elevation difference of 300 meters (**Fig. 1**). The selected site was a small forest patch of approximately 10,000 m^2 bordered by two small streams immersed in pine plantations, which prevents connectivity with other patches.

Capture-Mark-Recapture (CMR)

We will used the capture-mark-recapture (CMR) method in MARK software (White, 2014) with a closed population model (Williams et al. 2002) to determine the population parameters of *A. aff tolimensis*. Recaptures have been determined by the use of visual implant elastomer tags (VIE) (Anholt et al. 1998) (**Fig. 5**). Both seasons (wet and dry) have been explored, however, there were apparently no changes. We make random time-standardized transects (five hours each, 7:00 - 12:00) using the visual encounter survey method (Crump & Scott, 1994) within a small forest patch of approximately 10.000 m2. *A. aff tolimensis* males were determined by engaging in reproductive activities such as transporting tadpoles or by males that were actively calling (Duellman, 1966). Likewise, females were determined by engaging in reproductive behavior with males calling and juveniles through observations of small snout-vent length and low weight (**Fig. 4**, **8**). Thus, we hope to estimate apparent survival probabilities (Φ), recapture probabilities (p), and the realized population growth rate (l) for each interval between capture occasions, as a function of the adult class, time, season, and SVL acting additively (no interaction effects) in models including more than one variable per parameter. We will use the Akaike Information Criterion (AIC) to evaluate the models (Akaike, 1987) which incorporate the uncertainty in the process of parameter estimation (Burnham & Anderson, 2004). Likewise, the Akaike information criterion will be corrected for a small sample size (AICc; Akaike, 1973; Burnham & Anderson, 2004). We will calculate model-specific Akaike weights and if more than one model is chosen, we will calculate weighted averages.

Phenotypical analysis

We recorded spontaneous advertisement call of ten *Andinobates aff tolimensis* (**Fig. 6**, **Table 1**), two *Nymphargus rosada* and one *Centrolene antioquensis* (**Fig. 16**) with an Audio-Technica AT897 microphone coupled to a boompole K-Tek KE-89CC in a ZOOM H6 recorder positioned at approximately 70 cm directly in front every male calling. Due to temperature dependence for metabolic regulation in ectotherms, we also recorded air, substrate, and body temperature with a Hygro-Thermometer RH101 Extech IR and Klein tools IR5 dual laser infrared thermometer (**Fig. 9**). For spectral and temporal analysis, we used Raven Pro v1.6.3 (Center for Conservation Bioacoustics, 2014) with 44.1 kHz and 16 bits. The spectral parameters were analyzed with a Fast Fourier transformation under the Blackman algorithm with 724 samples by a window, a frequency grid with 2,048 samples using the discrete Fourier transform algorithm (DFT), and default values for other parameters. Call parameters measured were specific to every species and some of them were call duration (CD), pulse number (P), peak frequency (PF), lowest frequency (LF), highest frequency (HF), and bandwidth (BW). All spectral parameters were

measured 10 decibels below peak frequency. A copy of these calls will be deposited at Fonoteca Zoológica (www.fonozoo.com).

Furthermore, we recorded the snout-vent length (SVL) measures with a Mitutoyo Absolute CD-6" CSX digital caliper (nearest 0.01 mm). We also weighed frogs with a pocket scale (nearest 0.01 g) (**Fig. 7**, **8**). Finally, some amphibians were euthanized with benzocaine, fixed in 10 % formalin, and stored in 70 % ethanol at the Colección Zoológica de la Universidad del Tolima.

All fieldwork was carried out in strict adherence to the guidelines for work with amphibians and reptiles (Beaupre et al. 2004) under permit of collect and management was granted to the Grupo de Herpetología, Etología y Eco-fisiología of the Universidad del Tolima by the ANLA agency (Resolución Nº 02252 de 2019).

Microenvironmental analysis

Six environmental temperatures data have been recorded in this project (**Fig. 12**). We have obtained hundreds of records of substrate, body, and environmental temperatures for many *Andinobates* (**Fig. 9**). The methodology for these records is indicated in the phenotypic analysis. However, we also recorded 3326 microenvironmental temperatures from six MX2201 HOBO data loggers. Two of them were left within the microhabitat used (burrows) by *A. aff tolimensis*, two around 1.5 m on the ground, and two recording underwater temperatures (**Fig. 10**, **11**, **12**, **Table 2**). We hope to maintain these data loggers in the middle- (5 years) and long-term (10 years), for future analysis of climatic change.

Genetic and Bd analysis

To date, we have twenty samples of *Andinobates aff tolimensis* from two different sites (**Fig. 10**) and eight of *Pristimantis tribulosus* and *Nymphargus rosada* (**Fig. 15**, **16**). DNA will be extracted using the QIAGEN kit at the Instituto de Ecología of the Universidad Nacional Autónoma de México (México). The mitochondrial cytochrome oxidase I gene (COI) will be amplified from museum and field tissue samples to verify the taxonomic status. The sequences will be assembled, edited, aligned, and visually reviewed, using MUSCLE (Edgar 2004) in Geneious v7.1.9 (Kearse et al. 2012). For phylogenetic analyses and genetic distance, we will a tree estimated with Bayesian inference in MrBayes (Ronquist et al. 2012) and maximum likelihood with RAxML (Stamatakis 2014). The genetic distance will be calculated with MEGA (Kumar et al. 2016), following the parameters of Amézquita et al. (2013). Bd samples will be taken with swabs and sent for commercial qPCR analysis. However, Bd samples have not yet been taken because the MW113 swabs have not been obtained in Colombia.

Environmental workshops

Due to the importance of sharing knowledge and research with the Padua community, a proposal was made to hold at least eight environmental education workshops at the Institución Educativa Juan XXIII (https://www.facebook.com/IeJuanXXIIIPadua/?locale=es_LA). However, the rector of the institution Dalila Uribe only allowed four workshops (**Fig. 13**). Moreover, to these workshops, surveys have been carried out to understand the community's perception of the species and its threats. On the other hand, during the workshops stickers have been distributed and practical exercises have been carried out in the analysis of temperature and bioacoustics data with the students of the 10th and 11th grades (**Fig. 13**). At the end of the project, a general workshop will be held with all the students to share the results and deliver posters.

Outputs and results

1. **Determine short-term population dynamics of *Andinobates aff tolimensis* of a probably new population with capture-mark-recapture methods**

After seven field trips, 392 records (**Fig. 2**) of *Andinobates aff tolimensis* individuals have been made with 42 recaptured individuals (**Fig. 3**), which indicates 10.71% recapture, after 28 capture events. The population consisted of 123 males, 42 females, 46 juveniles, and 180 individuals without determining the sex or stage of maturity (**Fig. 4**). Recapture determination was successful through the visual implant elastomer tags (**Fig. 5**), indicating this method can be perfectly replicated due to tags do not lose on time. Furthermore, suggesting that these individuals survive more than three months in the field (personal observations). The data suggest that juveniles, females, and males occupy the habitat heterogeneously (**Fig. 4**).

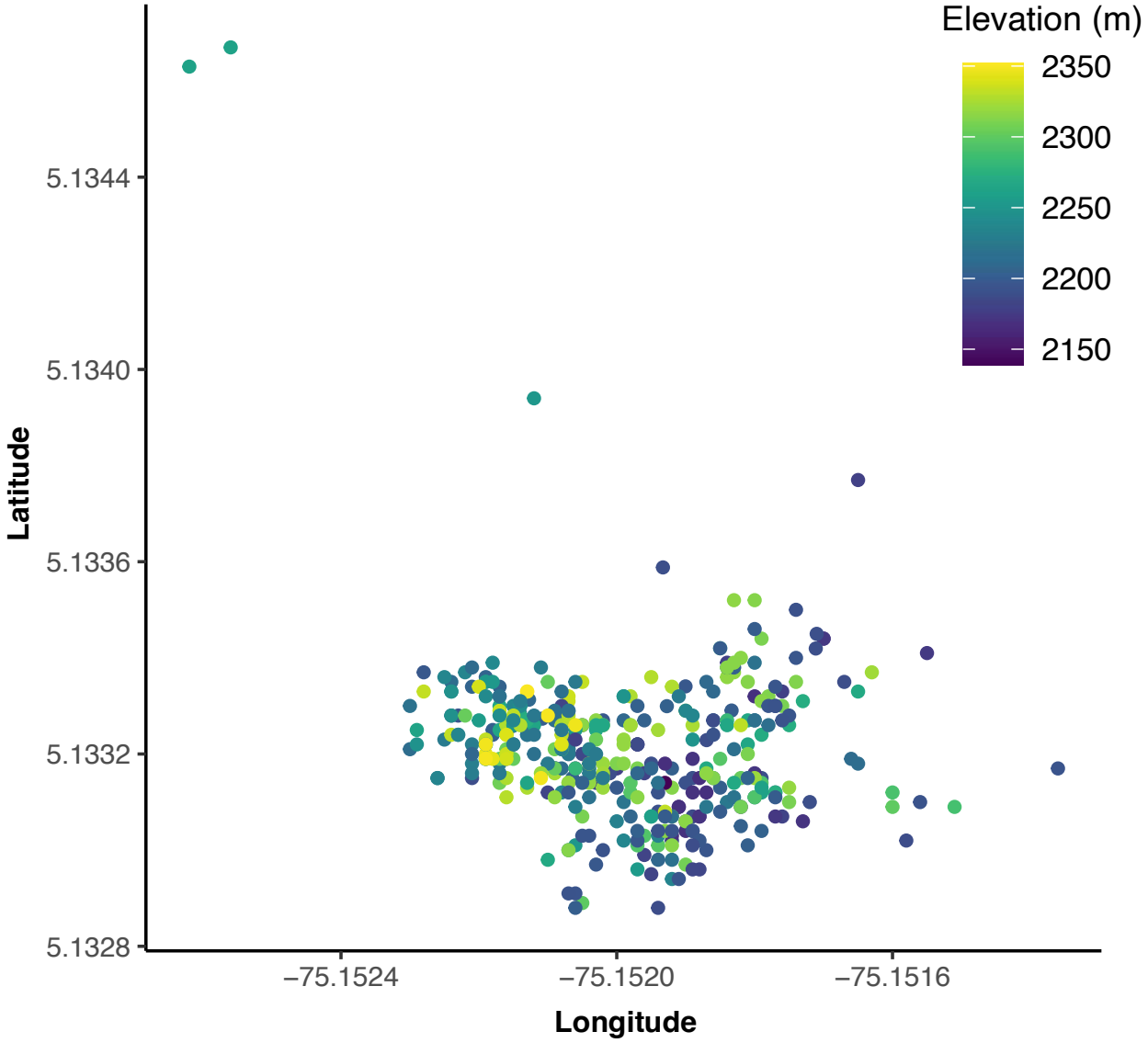


Figure 2. Spatial distribution of 392 records of *Andinobates aff tolimensis*.

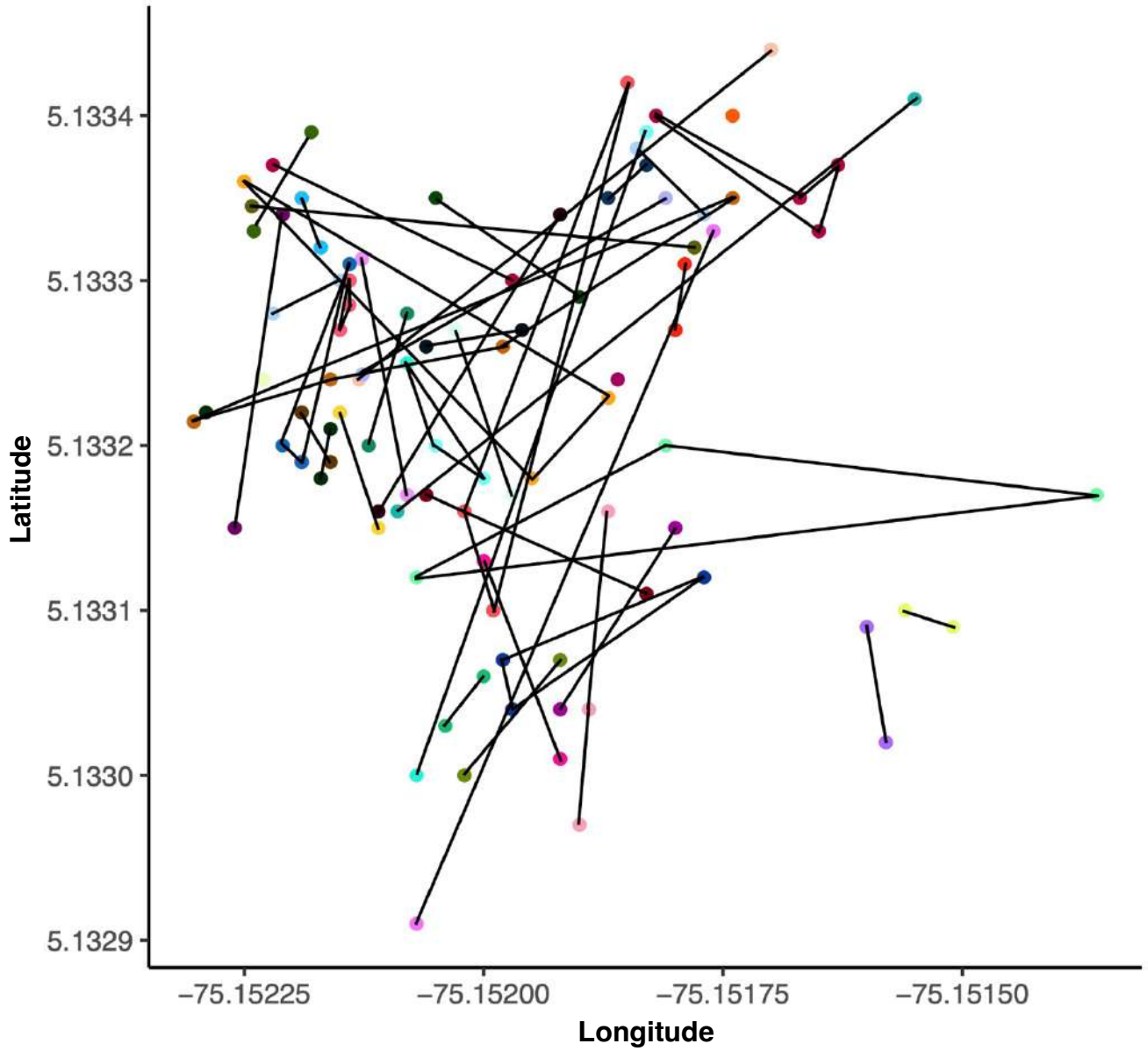


Figure 3. Spatial distribution of 42 individuals of *Andinobates aff tolimensis* capture-recaptures over four months. The colors indicate the spatial distribution of each capture and recapture. Points without connecting lines indicate individuals found at practically the same site.

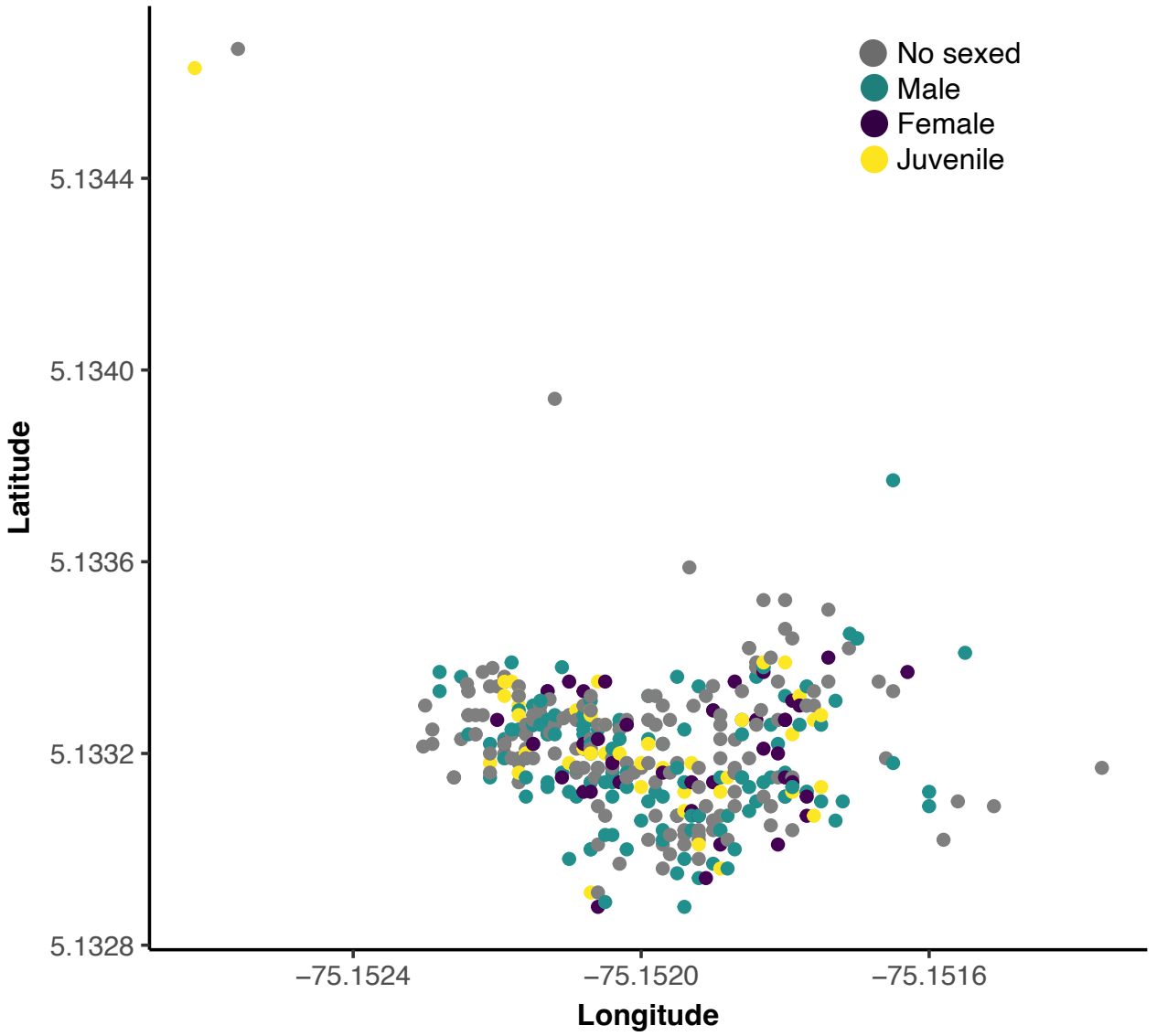


Figure 4. Spatial distribution of 392 sexed records of *Andinobates aff tolimensis*. 123 males (dark violet), 42 females (teal), 26 juveniles (subadults) (yellow), and 180 unsexed frogs (dark grey).

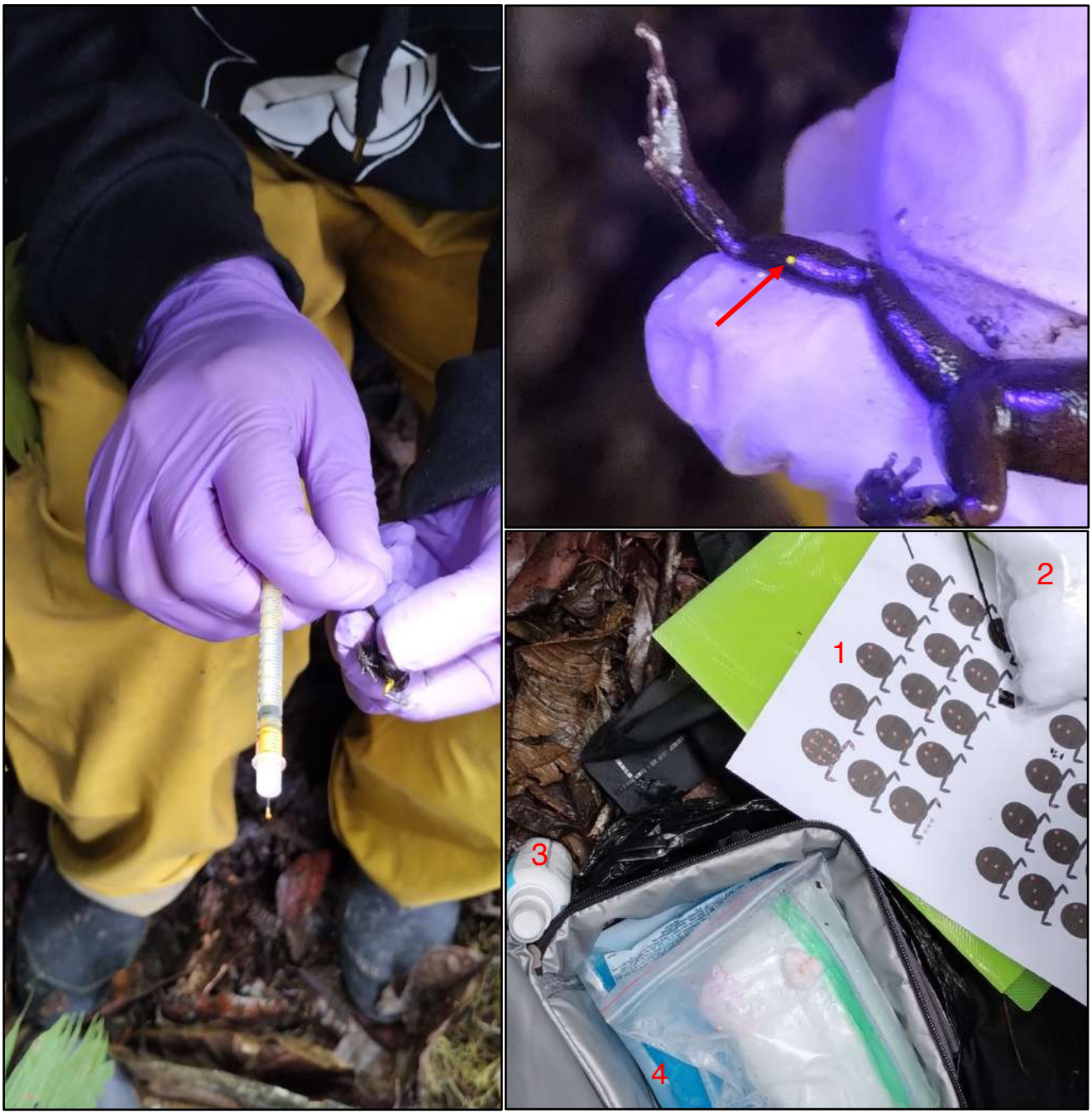


Figure 5. *Andinobates aff. tolimensis* individuals capture and marked with visual implant elastomer tags (VIE). Left, a frog of *A. aff tolimensis* handled to be marked with VIE. Top right, fluorescent orange marking of eleven frog under UV light. Bottom right, 1) index of marking tags, 2) cotton, 3) IXER 080 (chlorhexidine digluconate and excipients csp.) antiseptic solution to avoid infections of frogs and disinfect syringe needles with VIE, and 4) Ice pack for VIE transportation.

2. Determine temporal and spectral features of the calls of *Andinobates aff tolimensis*

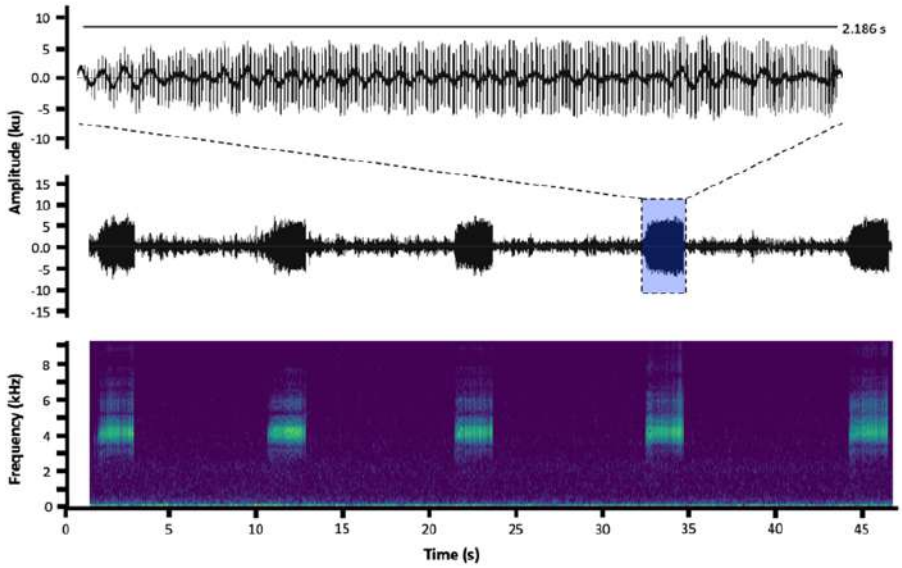


Figure 6. Call features of *Andinobates aff tolimensis***.** This call belongs to first individual recorded, who was recaptured on day three. The temporal and spectral features of the call are shown, as well as the duration. Analyses were made in Raven Pro v1.6. Analysis and results are presented in **Table 1.**

Table 1. Temporal and spectral parameters of the advertisement call of *A. aff tolimensis* of type locality (Falan) and the probably new population (Padua). Preliminary data are presented as the mean and range. NA = Not available. n = number of data.

Acoustic parameters	***Padua***	***Falan***
Call duration (s)	2.45 (1.26–3.38)	0.84–0.99
Pulses	148 (76–198)	NA
Dominant frequency (kHz)	4.47 (3.82–4.49)	4.73–5.22
Calls analyzed (n)	41 (seventeen frogs)	6 (one frog)
Size parameters		
Male mean SVL (mm)	17.86 (15.88–19.22)	17.87 (17.66–18.02)
Female mean SVL (mm)	19.66 (18.98–20.82)	18.28 (17.82–18.91)
Source	Our data	Bernal et al. 2007

3. Determine the morphology of *Andinobates aff tolimensis*

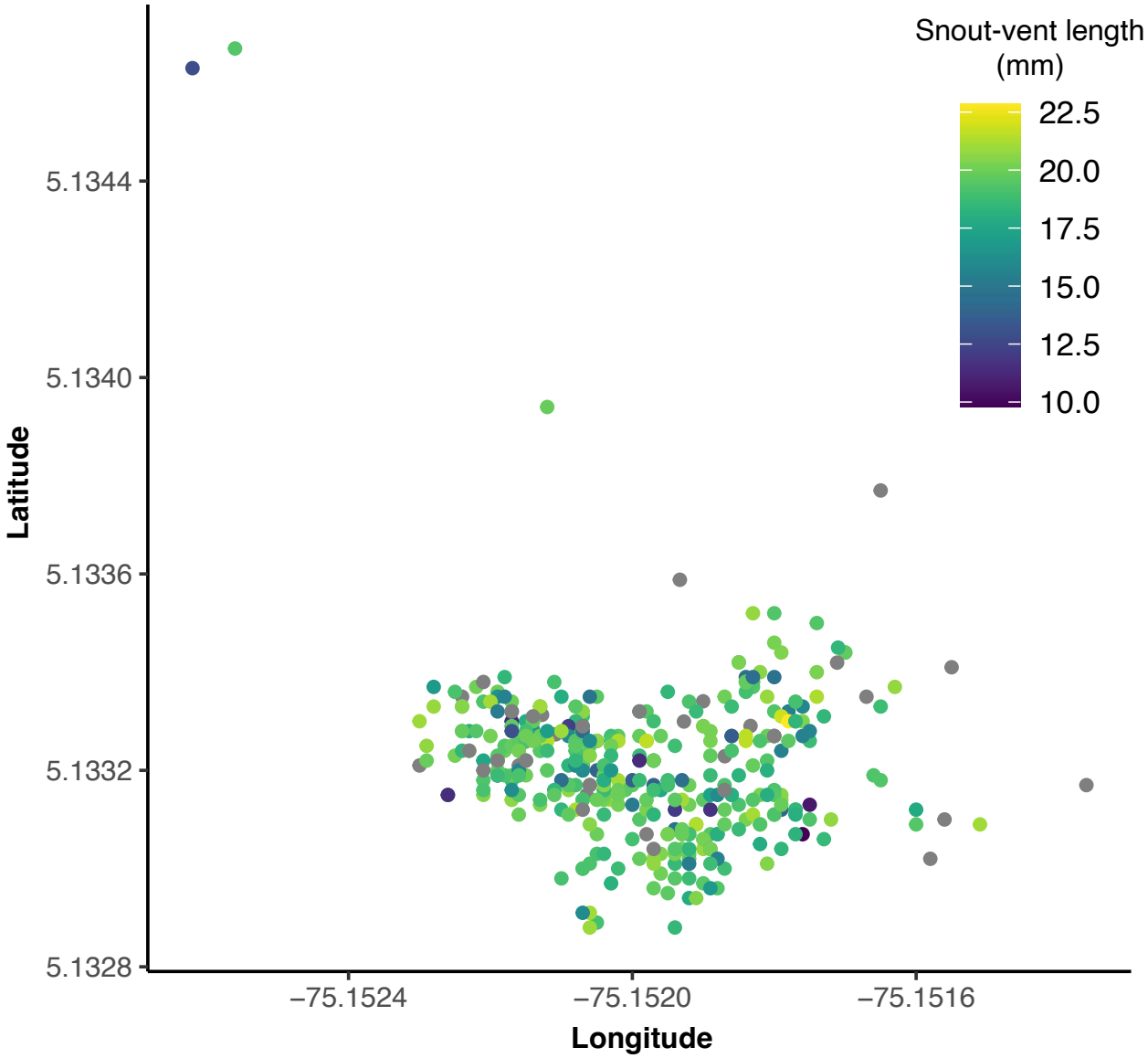


Figure 7. Spatial distribution of the body size of *Andinobates aff tolimensis*. Gray color indicates not measured frogs.

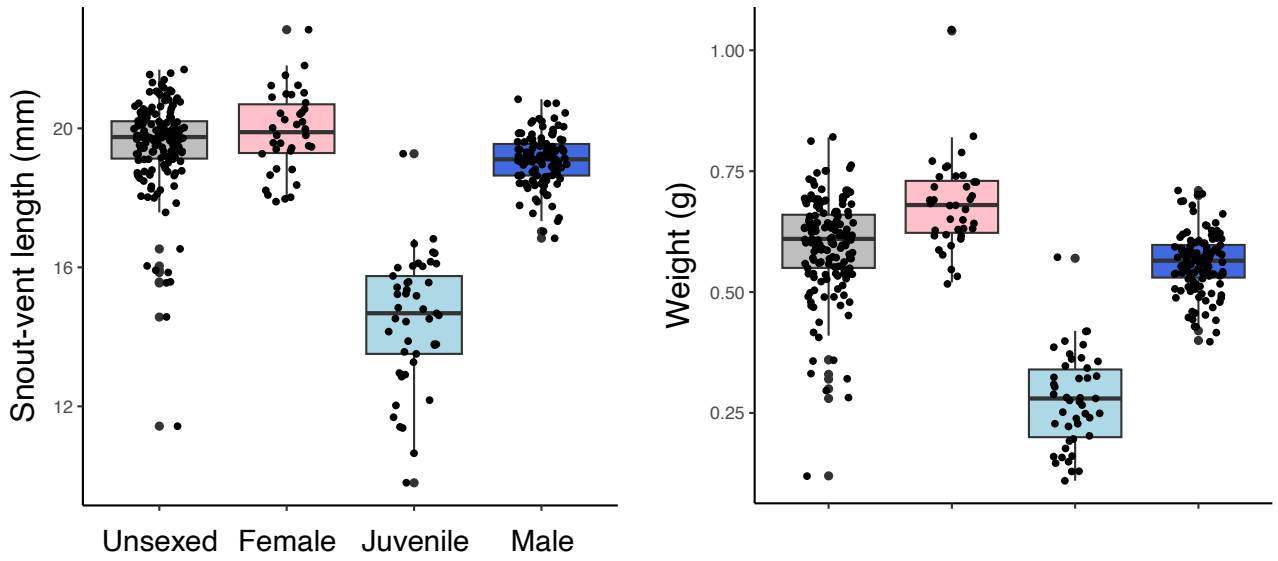


Figure 8. Differences in body size (left) and weight (right) of the different sexes and stages of maturity of *Andinobates aff tolimensis* population in Padua village. The relationship shows differences with females bigger than males.

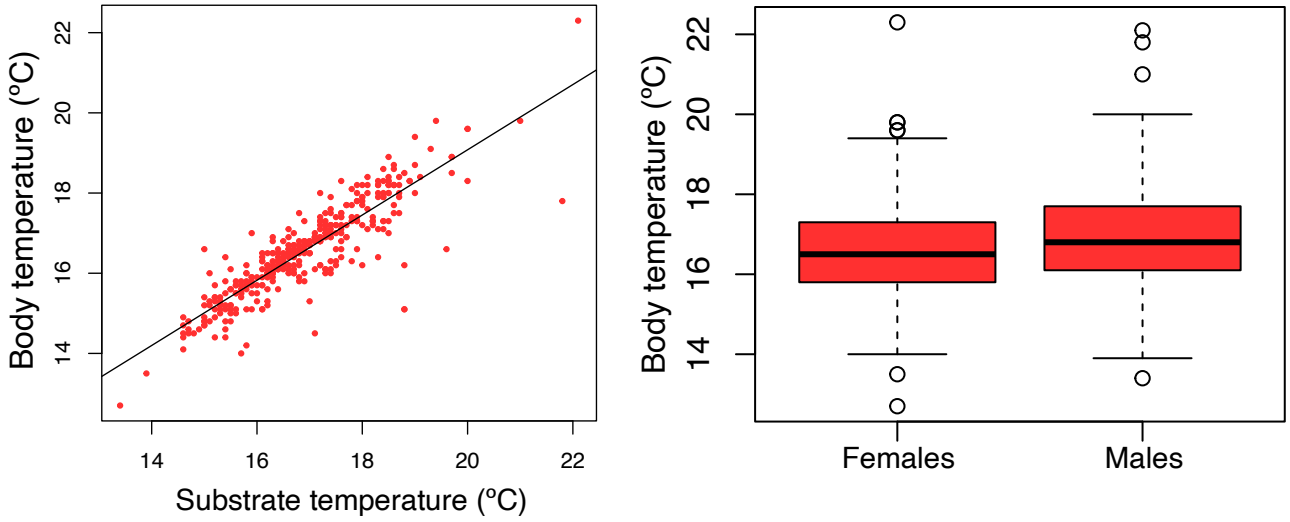


Figure 9. Relationship between environmental and body temperatures (left) and between sexes (right) for the population of *Andinobates aff tolimensis* in Padua village.

4. Contrast new and published sequences of mitochondrial genes to determine the status of a probably new population of *Andinobates aff tolimensis*

Due to the molecular biology laboratory of the Universidad del Tolima currently undergoing renovations, genetic analyzes cannot be carried out there. However, an export permit is being processed to take the tissues to the Instituto de Ecología of the Universidad Nacional Autónoma de México (México). There, the DNA extraction process will be carried out and later the sequence will be sent to Macrogen.Inc (South Korea). This objective has not started, but its delay was contemplated

since it is necessary first to obtain the largest amount of tissue to analyze the populations. Thus, it is expected that in a few days, the plate will be sent to Mexico and that it will already have the export permit.

5. Monitor microenvironment temperatures

Our data loggers (HOBO MX2201) have been recording temperature data each hour from February 23. We have left six data loggers, three in the population where population dynamics is being analyzed at 2350 masl (high site) and three in a small patch at 2085 (low site) (**Fig. 10**). The environments choose in each population were inside plastic containers with water (two data loggers, high and low sites). These containers were evaluated by visual observations of *A. aff tolimensis* tadpoles deposited by males (**Fig. 11**). Two data loggers at 1.5 m above the ground and two left inside burrows occupied by frogs. The data collected are shown in the **Fig. 12** and **table 2**. On the other hand, the water temperature data logger at the low site was lost, therefore, the data observed in **Fig. 12** and **Table 2** are incomplete. Due to this, two more data loggers were purchased and one left again at this site. These data will be fundamental for monitoring long-term environmental temperatures and also for future experiments on thermal tolerances in *Andinobates*.

Figure 10. Spatial distribution of six temperature data loggers. The dark green land cover represents Pinus crops, so the landscape shows the heavy impact of the loss of habitat for *Andinobates* frogs.

Figure 11. Water temperature recording from two data loggers. Top left, male of *Andinobates aff tolimensis* depositing tadpole in a plastic container at the low site (N5.12783, W-75.14875; 2085 masl). Top Right, plastic container with a tadpole at the high site (N5.12780, W-75.14877; 2350 masl), the picture was taken in May 30. Middle, metamorph from the low site in 42 develop stage based on Gosner

1960 (picture taken June 21). This metamorph was monitored from February 25. Bottom, HOBO MX2201 data logger inside plastic container at the low site.

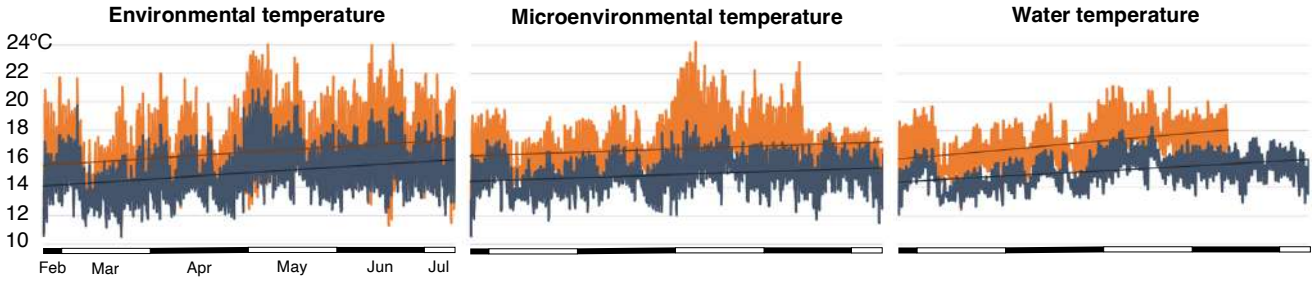


Figure 12. Temperatures recorded for the habitat of *Andinobates aff tolimensis*. Left, temperatures recorded around 1.5 meters above ground. Middle, temperatures recorded within burrows inhabit by *A. aff tolimensis* frogs. Right, temperatures recorded inside plastic containers with water occupied with tadpoles of *A. aff tolimensis* frog (**Fig. 11**). The HOBO MX2201 data loggers recorded 3.325 hours from February 23 to July 11 of 2023. Scales are standardized. Orange indicates three data loggers at the low site (environment N5.12783, W-75.14869; microenvironment N5.12783, W-75.14875; water N5.12789, W-75.14864) and blue gray three data loggers at the high site (environment N5.12780, W-75.14877; microenvironment N5.13327, W-75.15192; water N5.12780, W-75.14877). The maximum, mean, and minimum temperatures recorded of each data loggers are shown in Table 2. The water temperatures at the low site (orange) were only taken through June 14 because the data logger was lost and possibly stolen.

Table 2. Summary of temperatures recorded by the data loggers. The population at the low site experiments at higher temperatures than the high site population, even up to nearly six degrees of maximum temperature in the microenvironmental temperatures with a small elevation difference (265 m). Temperatures are in degrees Celsius (°C).

	Environmental temperature		**Microenvironmental temperature**		**Water temperature**	
	Low site (2085 masl)	**High site (2320 masl)**	**Low**	**High**	**Low**	**High**
Mean	16.44 ±2.34	15.01 ±1.70	16.71 ±1.74	14.92 ±1.23	17.02 ±1.49	15.17 ±1.08
Minimum	10.77	10.51	12.05	10.59	12.40	12.10
Maximum	24.06	20.89	24.24	18.70	21.10	18.19

6. Evaluate the presence of *Batrachochytrium dendrobatidis* (bd)

This has not yet been developed due to the high costs of importation of MW113 swabs since they could not be obtained in Colombia. We are working to find some strategy, such as an academic collaboration that allows us to complete this objective.

7. **Promote environmental education with students and different stakeholders, involving them in field trips and environmental workshops.**

Although it was proposed to hold workshops with the students during each field trip, there was only permission from the Rector Dalila Uribe to develop four (amphibian taxonomy, ecosystem services, bioacoustics, and climate change, loss of biodiversity, pandemics, and panzootic) (**Fig. 13**). On the other hand, due to the difficulty in accessing the field site, we did no work with students. However, in some workshops live and collected frogs were brought so that the students knew the species. Likewise, a small field trip was made to take temperature data and record songs and calls, with these data, a bioacoustics workshop was held with students from the last grades (10 and 11) (**Fig. 13**).

Figure 13. Environmental workshops at the (Institución Educativa Juan XXIII (https://www.facebook.com/IeJuanXXIIIPadua/?locale=es_LA) (N5.13278 W-75.14379). Top left, workshop on amphibians with children from the 5th grade. Top right, stickers that have been delivered out during some workshops with students. These stickers have also been delivered to different small-

business premises and to the general public. Middle, bioacoustics workshop with students of 11th grade. Bottom, stickers given to children during an event in the main park.

Other outputs and results

Figure 14. Socialization of study for the construction of a new highway in Padua. At the end of June, we were invited by the Padua community to participate in the socialization of a new highway to be built in Padua. Although it was not possible to know exactly the properties that will be intervened, this infrastructure development could be a new pressure for the already diminished population in the future. Therefore, we will be attentive to future conversations and monitor to avoid the loss of the population of *Andinobates* or other threatened amphibians.

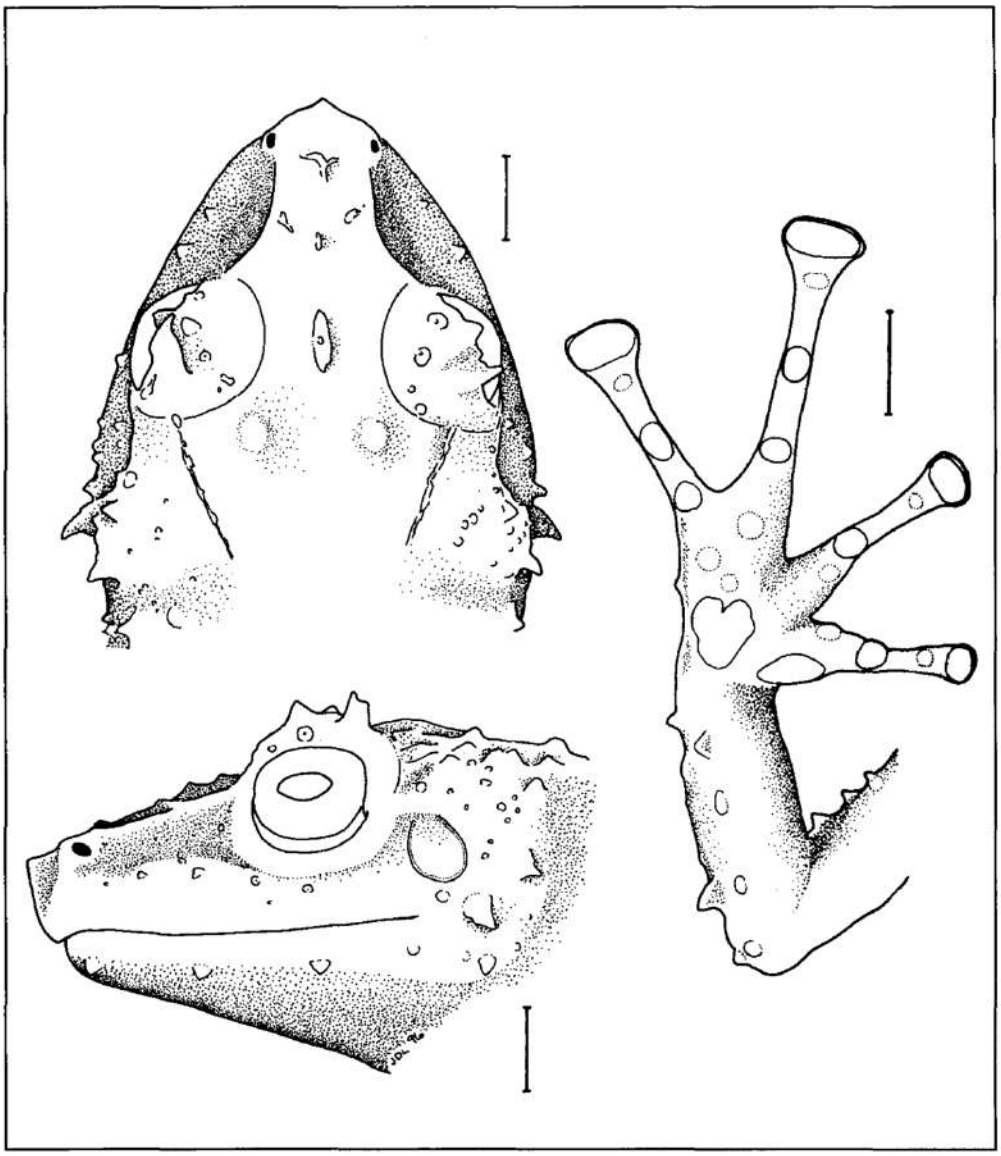


Figure 4. *Eleutherodactylus tribulosus* sp. nov. (ICNMHN 37169). Scales equal 2 mm.

Figure 15. Thorny rain frog (*Pristimantis tribulosus*) found in the study area of *Andinobates aff tolimensis*. We are currently preparing a report with the first record of *P. tribulosus* outside its type locality. This species since it was described in 1997 has only been reported in the Parque Nacional Natural Selvas de Florencia, in addition, it is Critically Endangered (IUCN, 2017). Top, morphological scheme of the original description (Lynch & Rueda-Almonacid, 1997). Bottom, two females of *P. tribulosus* with morphological type characters.

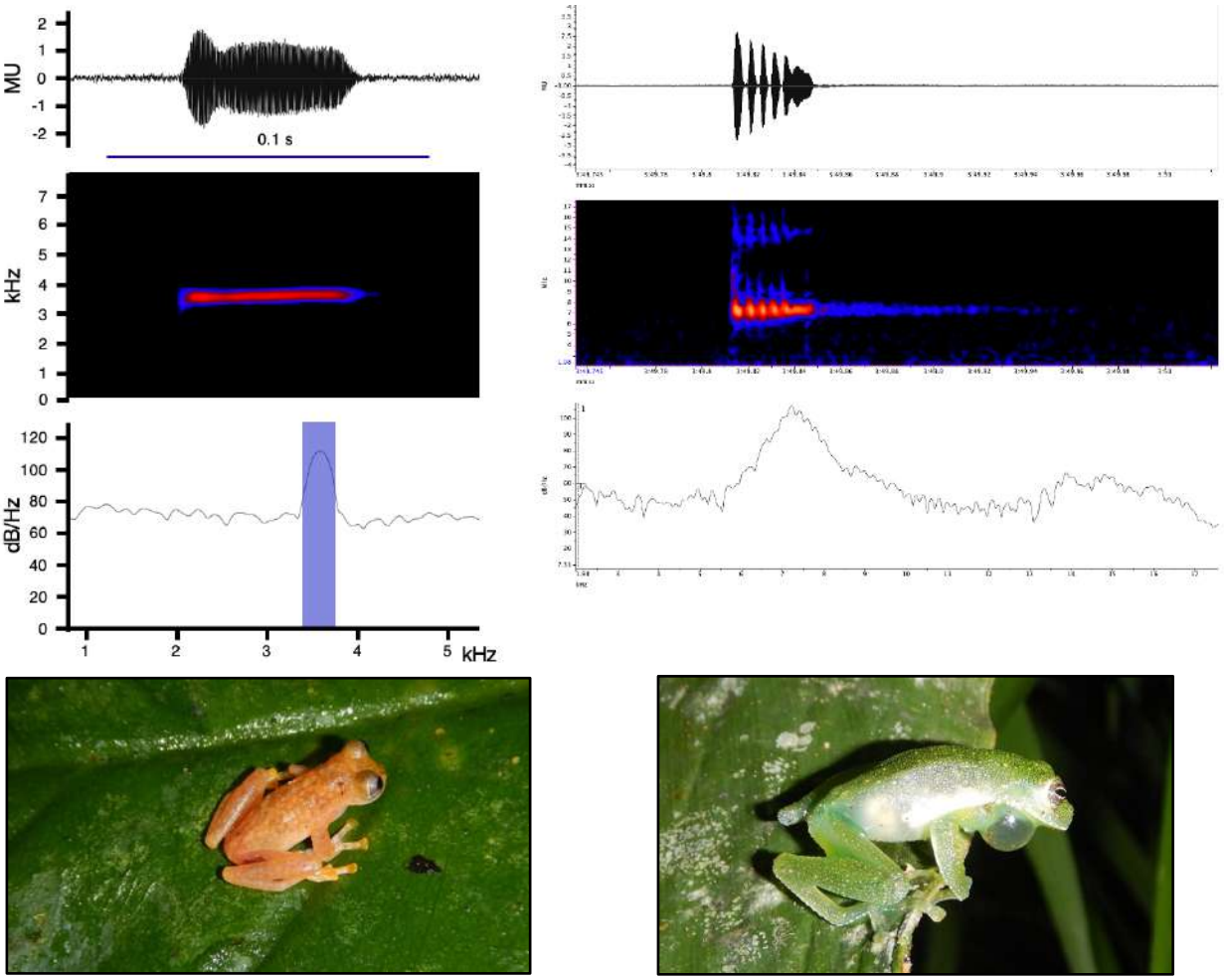


Figure 16. Recorded calls of other frog species found. Left, call of *Nymphargus* rosada (N5.11920, W-75.15069; 2164 masl). Right, call of *Centrolene antioquensis* (N5.13531, W-75.15353; 2407 masl). These data are being prepared for a joint publication with the record of *Pristimantis tribulosus* (**Fig. 15**).

Figure 17. *Andinobates aff tolimensis* with serious injury marks. Record A384 from July 13, 2023 is a recapture of a juvenile individual A214 captured on May 31, 2023. This record is important since it shows the individual heavily lacerated, probably as a result of an attempt to be preyed on.

Figure 18. Habitat loss for *Andinobates aff tolimensis*. Left, view from Padua village. The highlight shows the small patch where the demography study is done. Right, view from the small patch. The large extensions of introduced pine (*Pinus patula and Pinus maximinoii*) crops are highlighted.

Financial report

To date, all equipment (data loggers and visual implant elastomer tags) were purchased. However, since one data logger was lost, we bought two more to replace this one and eventually in case we lost another. On the other hand, all the equipment provided by the Universidad del Tolima has worked perfectly and no extra expenses have been generated. However, the costs of the elastomers and data

loggers were higher due to the devaluation of the Colombian peso, shipping, customs, and taxes. Costs for genetic and Bd analysis have not yet been incurred.

Description	Cost	Spent money	Remaining money
Lodge	860	690	170
Stickers (1500) and posters	200	175	25
Batrachochytrium dendrobatidis qPCR test	380	0	380
Food	1532	1200	332
Ten round trips	766	610	156
Sanger sequencing	446	0	446
DNeasy Blood & Tissue Kit (100)	510	0	510
Visual Implant Elastomer kit (6ml)	470	495	-25
Batteries charger and batteries	119	0	119
Ten Brady Yellow Flagging	54	0	54
Six Hobo pendant MX2201 temperature data loggers	408	542	-134
Nitrile gloves, masks, ziploc bags, ethanol, hipoclorithe, Eppendorf tubes	213	213	0
Internet and cellphone	42	35	7
Two Hobo pendant MX2201 temperature data loggers	187.94	187.94	0
	6000	**4147.94**	**1852**

Figure 19. Preliminary financial report shows the initial budget, spent money, and remaining money. Negative values indicate higher equipment costs. The red sentence indicates cost of two new data loggers bought to replace one data logger lost (**Fig. 12**).

Conclusion

All our objectives have been partially met. First, we were able to record the focal species, this is important given that it is a threatened species. Furthermore, we were able to understand a little better the ecology and behavior of the species. For example, *Andinobates aff tolimensis* occurs in an agricultural matrix with highly fragmented (**Fig. 1**, **10**, **18**). The species is relatively easy to sample, with a recovery rate of 10.7% and about 400 records. Unfortunately, despite not having the demographic analysis yet, we can presume that the population will be quite small. Thus, connectivity measures for the populations of *A. aff tolimensis* are urgent, due to the strong loss of habitat. Likewise, we were also able to record the call, body measures, body temperatures, and microenvironmental temperatures.

We have not yet performed the genetic analyzes but we hope to have these data in the coming months and even already have a manuscript submitted. On the other hand, we are already writing a manuscript on newly recorded threatened species, one of the most important results so far.

Fine microenvironmental data are scarce for threatened or non-threatened species, here we highlight that our data indicate that the microhabitat of *A. aff tolimensis* (burrows) is a temperature buffer that reduces maximum temperatures by up to 6 degrees (**Fig. 12**, **Table 2**). The microenvironmental results suggest that the use of the microhabitat in *A. aff tolimensis* is important when it comes to compensating for the temperature changes that can be recorded in the habitat, more so as this is a diurnal species.

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