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Conservation of the critically endangered frog
Telmatobufo bullocki in fragmented temperate forests
of Chile

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Abstract

Amphibians are currently facing several threats and are suffering severe population declines and extinction worldwide. *Telmatobufo bullocki* (Anura: Calyptocephalellidae) is one of the rarest and most endangered amphibian species in Chile's temperate forests. It is the fifth most evolutionarily distinct and globally endangered (EDGE) amphibian in the world, and one of the world's top 100 priority species for conservation (Zoological Society of London, 2011). This stream-breeding frog is micro-endemic to the coastal Nahuelbuta mountain range in central-south Chile (37°-38°50' S), a hot-spot for conservation. This area has suffered severe loss and fragmentation of native forest, which has been replaced by extensive commercial plantations of exotic pines and eucalyptus. Despite its potential detrimental effects, the impact of native forest loss on this species has not been studied before. Furthermore, few historical observations exist, and the ecology and behaviour of the species is poorly known. In addition, current status and location of extant populations are uncertain, which makes conservation and targeted habitat protection difficult.

Through the use of different approaches and modern conservation tools this thesis aims to make a significant contribution to the conservation of *T. bullocki* and its habitat. Historical and new locations were surveyed to identify extant populations. A distribution modeling approach (i.e. Maxent) was used to infer the species' distribution within Nahuelbuta, generate a predictive habitat suitability map, identify important environmental associations, and assess the impact of main environmental threats (i.e. native forest loss, climate change). Field-based research (e.g. surveys, radio-tracking) was done to extend the

ecological and behavioural knowledge of the species (e.g. movement patterns and habitat use), and identify critical aquatic and terrestrial habitat for protection (i.e. core habitat). Mitochondrial and specifically developed microsatellite genetic markers were used to measure levels of intra-specific genetic variability, define genetic population structure and connectivity, infer evolutionary history (phylogeography), estimate effective population size and detect demographic changes (e.g. bottlenecks). Finally, a landscape genetics approach was used to relate landscape characteristics to contemporary patterns of gene flow, and identify important landscape features facilitating (i.e. corridors) or hindering (i.e. barriers) genetic connectivity between populations.

Telmatobufo bullocki was found in nine basins within Nahuelbuta, including historic and new locations. Presence of *T. bullocki* was positively related to the amount of native forests in the landscape. However, some populations persist in areas dominated by exotic plantations. Some frogs were found living under mature pine plantation adjacent to native forest, but no frogs were found in core plantation areas. *T. bullocki* makes extensive use of terrestrial habitat adjacent to breeding streams during the post-breeding season, moving up to 500 m away from streams. A core terrestrial habitat of at least 220 m from streams is proposed for the protection of populations. Population genetics and phylogeography revealed significant population structure. The northernmost and disjunct population of Chivilingo is geographically and genetically isolated from all other sampled populations and was identified as a separate evolutionary significant unit (ESU). The population of Los Lleulles was also identified as a separate management unit, while the remaining populations were grouped into two clusters forming a larger and more connected meta-population. Connectivity within groups was high, suggesting individuals are able to

disperse between neighbouring basins. Levels of genetic diversity were not homogeneous, and were lowest at Los Lleulles and highest at Caramávida. Results suggest disjunct populations are at highest risk and should be prioritised for restoration and habitat protection, while management of meta-populations should aim at maintaining and improving connectivity among basins. Landscape genetic results identified streams and riparian habitat as dispersal pathways, and least-cost-path analysis was used to identify a potential connectivity network.

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List of Acronyms and Abbreviations

Acronym	Meaning
AIC	Akaike Information Criterion
Bd	<i>Batrachochytrium dendrobatidis</i>
BI	Bayesian Inference
BIC	Bayesian Information Criterion
BSP	Bayesian Skyline Plot
BU	Butamalal
CA	Caramávida
CH	Chivilingo
CL	Calebu
COI	Cytochrome c oxidase subunit 1
CONAF	Corporacion Nacional Forestal de Chile
CR	Critically endangered (IUCN threat category)
CY	Cayucupil
CWD	Coarse woody debris
DD	Data Deficient (IUCN threat category)
DO	Dissolved oxygen
E	Elevation
EDGE	Evolutionarily distinct and globally endangered
EM	Expectation maximization algorithm
ESS	Effective sample size

ESU	Evolutionarily significant unit
FSC	Forest Stewardship Council
HCVA	High Conservation Value Area
HKY+I	Hasegawa, Kishino and Yano model of DNA substitution with invariable sites
HPD	Highest posterior density
HSI	Habitat suitability index
HU	Huilquehue
HWE	Hardy-Weinberg equilibrium
IAM	Infinite Allele Model
IBD	Isolation by distance
IBR	Isolation by resistance
IUCN	International Union for Conservation of Nature
LC	Land cover
LCP	Least-cost path
LL	Los Lleulles
ML	Maximum likelihood
MW	Mega Watt
mya	Million years ago
Ne	Effective population size
NR	Nahuelbuta Range
PCR	Polymerase chain reaction
PR	Provoque
SC	Specific conductance

SD	Standard deviation
SE	Standard error
SI	Slope
SSD	Sum of square deviations
St	Proximity to stream
SVL	Snout-to-vent length
TMRCA	Time to most recent common ancestor
VES	Visual encounter surveying
VIE	Visible implant elastomer

