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Research Article

Habitat Suitability Modelling of *Rhacophorus reinwardtii* (Schlegel, 1840) in Java, with Notes on Habitat Characteristics from Malang Region-East Java

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ABSTRACT
The distribution of the Reinwardt's Gliding Frog, *Rhacophorus reinwardtii* (Schlegel, 1840), was recently thought to be restricted in Java. Furthermore, the constant and rapid deforestation in Java highlights the significance of the frog's habitat requirements in establishing the accurate species' conservation status. This study aims to predict the suitable habitat for this species in Java and to identify the habitat characteristics in Malang Region-East Java. Using the Maximum Entropy (MaxEnt) approach, we modelled the distribution of species and examined numerous types of breeding sites in Malang Region. Our findings revealed that the species inhabits a much more restricted and fragmented habitat in Java, where it inhabits both forest and agroforestry areas.

INTRODUCTION

Reinwardt's Gliding Frog, *Rhacophorus reinwardtii* (Schlegel, 1840) was previously thought to be widely distributed in Java, Sumatra, the Malay Peninsula, and Borneo (Ohler & Delorme 2006). Research has found that populations outside of Java Island are classified as other species: population from the Malay Peninsula and Southern Thailand are classified as *R. norhayatiae* Onn and Grismer, 2010; population from southern Vietnam are classified as *R. helenae* Rowley, Tran, Hoang, and Le, 2012; population from Sabah and Sarawak are classified as *R. borneensis* Matsui, Shimada, and Sudin, 2013. Several discovery of new species indicates that R. reinwardtii is restricted to Java (Frost 2021).

R. reinwardtii is found at elevations ranging from 20 to 1800 m asl (above sea level) on Java Island (Kurniati 2008; Yazid 2009; Ratna & Wijaya 2013; Wening 2017; Septiadi et al. 2018; Amrullah 2019; Setiawan et al. 2019; Kadafi et al. 2019; Malik 2019; Deslina 2021). It was discovered in a previous study that *R. reinwardtii* can live in both primary and secondary tropical rainforests (IUCN Red List of Threatened Species 2022). In addition, there are few reports of *R. reinwardtii* in production forest areas or residential areas near forest boundaries (Iskandar 1998; Ohler & Delorme 2006). Aside from that, the rare occurrence of *R. reinwardtii* at low-to-moderate altitudes is also questionable, thereby, it is difficult to determine if this phenomenon is due to a lack of data or a shift in the suitable altitudinal range. Between the 19th and 20th centuries, the state of the forests in Java Island changed dramatically (Peluso 1991). Currently, this has potentially and negatively affect *R. reinwardtii* survival in its natural habitat in Java.

The assessment of the conservation status based on The IUCN Red List of Threatened Species (2022) classifies *R. reinwardtii* as a species of least concern due to its widespread distribution, presumed large population, and lack of data indicating a significant population decline. Nonetheless, it is essential to analyze and reevaluate the distribution using alternative methods, such as species distribution modelling. In addition, information on the habitat requirements of *R. reinwardtii* is lacking. In this study, we aim to predict the suitable habitat for this species in Java and to identify the habitat characteristics for this species in Malang Region. This will hopefully help to shed a light on the current status of *R. reinwardtii* and support its action plan and conservation strategy in the future.

MATERIALS AND METHODS

Study area

Java is a 12829.7 ha–island that serves as the social, economic, political, and cultural center of Indonesia (Whitten et al. 1996; BPS 2015). The island has a variety of land cover types, including forested areas, lowland cultivation, regrowth, grassland, savanna grassland, unvegetated land, and land used primarily for the settled garden, arable, or tree crop cultivation (Whitten et al. 1996). Since 2001, Java has concerningly experienced up to 264.7 ha of deforestation (150 ha in East Java; 75.9 ha in Central Java; 38 ha in West Java) (Global Forest Watch 2021).

East Java, recognised for its concerning deforestation rate, specifically in the Malang Region, presents a convincing case study. The Malang Region, including Malang City, Malang Regency, and Batu City, is a metropolitan area located in the province of East Java, Indonesia. It is primarily known for its urban and suburban areas situated in highland scenery, although its southern area is characterised by lowland topography. The geographical area in concern is in close proximity to a number of mountain ranges, including but not limited to Mt. Bromo-Tengger-Semeru to the east, and Mt. Arjuno-Welirang-Anjasmoro and Mt. Panderman-Kawi-Butak to the west.

Species distribution modelling

The Species Distribution Model (SDM) uses maximum entropy (MaxEnt) to produce relevant and robust result even with limited occurrence records (Proosdij et al. 2016). The minimum distance between two closely occurring points should be more than 1.5 km since the predictor variables resolution is 30 arc-second (~1 km^2). A total of 23 occurrence points of *R. reinwardtii* (datum: WGS 84) from western to eastern Java were utilised after the filtering process. These points were acquired through direct observation (6 occurrences) and literatures survey (17 occurrences) (Kurniati 2002; Kurniati 2003; Yazid 2009; Yanuarefa et al. 2012; Ratna & Wijaya 2013; Mumpuni 2014; Wening 2017; Amrullah 2019; Kadafi et al. 2019; Malik 2019; Setiawan et al. 2019; Idrus 2020).

The predictor variables was selected based on the species biology and ecology which included climatic, topographic, and biophysics predictors. A total of 24 independent predictor variables with 30 arc-second resolution were employed, these include 19 bioclimatic variables, representing seasonality, extremity, and annual trend from Worldclim v2.1 datasets (https://worldclim.org/data/worldclim21) (Fick & Hijmans 2017);

digital elevation modeling data datasets (http://srtm.csi.cgiar.org) (Jarvis et al. 2008), which were derived into slope data and roughness data; Landsat cloud-free image datasets composite (https://glad.earthengine.app/view/global-forest-change) (Hansen et al. 2013) which were extracted for Normalised Difference Vegetation Index (NDVI); and tree canopy cover data for the year 2000 datasets (Hansen et al. 2013) (Table 1).

These predictor variables were reprojected, resampled, and clipped accordingly to the study area. The preparation of predictor variables was carried out in RStudio v.3.4.1 (R Core Team, 2013), using various packages including *rgdal* (Bivand et al. 2022), *rgeos* (Bivand & Rundel 2022), *sp* (Pebesma & Bivand 2005), *raster* (Hijmans 2022), and *tidyr* (Wickham & Girlich 2022).

Prior to the SDM analysis utilising MaxEnt, it is crucial to correct for multicollinearity and exclude highly correlated predictor variables to prevent overfitting prediction (Merow et al. 2013). Consequently, we utilised a multicollinearity test with a variance inflation factor (VIF) to detect collinearity between variables using a stepwise procedure. The multicollinearity test for predictor variables was conducted in RStudio v.3.4.1 utilising multiple packages, i.e., *usdm* (Naimi et al. 2014) and *terra* (Hijmans 2022). This procedure eliminated 13 predictor variables due to collinearity issues.

The maximum entropy technique utilising the MaxEnt v. 3.4.1 software (https://biodiversityinformatics.amnh.org/open_source/maxent/) was utilized (Phillips et al. 2006). We jackknifed our model to determine the best predictor variables for the distribution of *R. reinwardtii*, used clog-log output format, and examined response curves. We used 10% of the data for testing and 90% of the data for training; a total of 15,000 background points were used, ten replications were performed

Table 1. Details of predictor variables employed in MaxEnt of *R. reinwardtii* in Java, using 30 arc-second resolutions.

Code	Predictor variables	Source	Unit
bio1	Annual mean temperature	WorldClim	ºC
bio2	Mean diurnal range	WorldClim	ºC
bio3	Isothermality	WorldClim	%
bio4	Temperature seasonality	WorldClim	ºC
bio5	Max. temperature of warmest month	WorldClim	ºC
bio6	Min. temperature of coldest month	WorldClim	ºC
bio7	Temperature annual range	WorldClim	ºC
bio8	Mean temperature of wettest quarter	WorldClim	ºC
bio9	Mean temperature of driest quarter	WorldClim	ºC
bio10	Mean temperature of warmest quarter	WorldClim	ºC
bio11	Mean temperature of coldest quarter	WorldClim	ºC
bio12	Annual precipitation	WorldClim	mm
bio13	Precipitation of wettest month	WorldClim	mm
bio14	Precipitation of driest month	WorldClim	mm
bio15	Precipitation seasonality	WorldClim	%
bio16	Precipitation of wettest quarter	WorldClim	mm
bio17	Precipitation of driest quarter	WorldClim	mm
bio18	Precipitation of warmest quarter	WorldClim	mm
bio19	Precipitation of coldest quarter	WorldClim	mm
trecov	Tree canopy cover	Hansen et al. 2013	%
ndv	Normalized difference vegetation index	Hansen et al. 2013	Nil
ele	Elevation	Jarvis et al. 2008	m
slo	Slope	derived from ele	°
rou	Roughness	derived from ele	°

using subsampling replication run type and 500 iterations were maintained (Khan et al. 2022); we used regularisation of 3 because it performs better and reduces the over-fitting of the prediction results. We reported the receiver operating characteristic (ROC) area, which indicated the area under curve value (AUC value). If the AUC value is greater than 0.70, it indicated that the predictive model is performing well (Araújo et al. 2005). We reported the average AUC value under the ROC curve and calculated the probability of presence based on the average of ten replicate models.

For the binary presence/absence map, we used the 10th percentile training threshold, which excluded all regions with habitat suitability lower than the suitability values for the lowest 10% of occurrence records. The binary presence/absence map was derived from the previous probability of presence output. This analysis was conducted in RStudio version 3.4.1, following Hu and Jiang (2018).

Using the Geospatial Conservation Assessment Tool (GeoCAT) (http://www.kew.org), we also determined the area of occupancy (AOO) and extent of occurrence (EOO) polygons from *R. reinwardtii* occurrence points.

We visualised the results with the QGIS 3.18 software and arranged three outputs: i) the species occurrence overlaid with tree canopy cover, ii) the probability of presence map, and iii) the binary presence/absence map.

Field observation

To illustrate the general habitat characteristics of *R. reinwardtii*, we investigated the previously documented species occurrence and breeding sites in Malang Region (Figure 1), as representative of Java. In addition, the occurrence in East Java may represent the actual threats to their populations, as East Java is the province with the highest extent of forest degradation (Global Forest Watch 2021).

From August 2021 to February 2022, we conducted the Visual Encounter Survey in conjunction with the Breeding Site Survey (Heyer et al. 1994), and collected data on habitat characteristics. Each of these activities is conducted on every month throughout the observation period. Observations were conducted during the time period of 18:00 to 22:00 to account for the peak activity exhibited by amphibians. After documenting the individuals encountered and the breeding site, measuring the microclimate and abiotic factors and conducting a comprehensive examination of the individuals and breeding sites, all captured individuals were released back into the habitat where they had been found. To avoid the degradation risk of breeding sites, the suffering of animals, and the transmission of animal disease (e.g., chytridiomycosis), we strictly adhere to the biosecurity aspect during the surveys (wearing gloves, disinfected the tools, be aware of the surroundings).

RESULTS

Species distribution modelling

Based on the selected remaining predictor variables (11 predictor variables: bio2, bio3, bio4, bio13, bio14, bio18, bio19, trecov, ndvi, ele, and slo), the MaxEnt models evaluation revealed that the selected predictor variables produced the best models for estimating the probability of *R. reinwardtii* presence. The averaged test omission rate and predicted area performed better than random when tested for omission and demonstrated good reliability of prediction (Appendix 1). The average AUC of the replicate tests is 0.878, and the standard deviation is 0.079, indicating that the SDM has a strong predictive performance (Appendix 2).

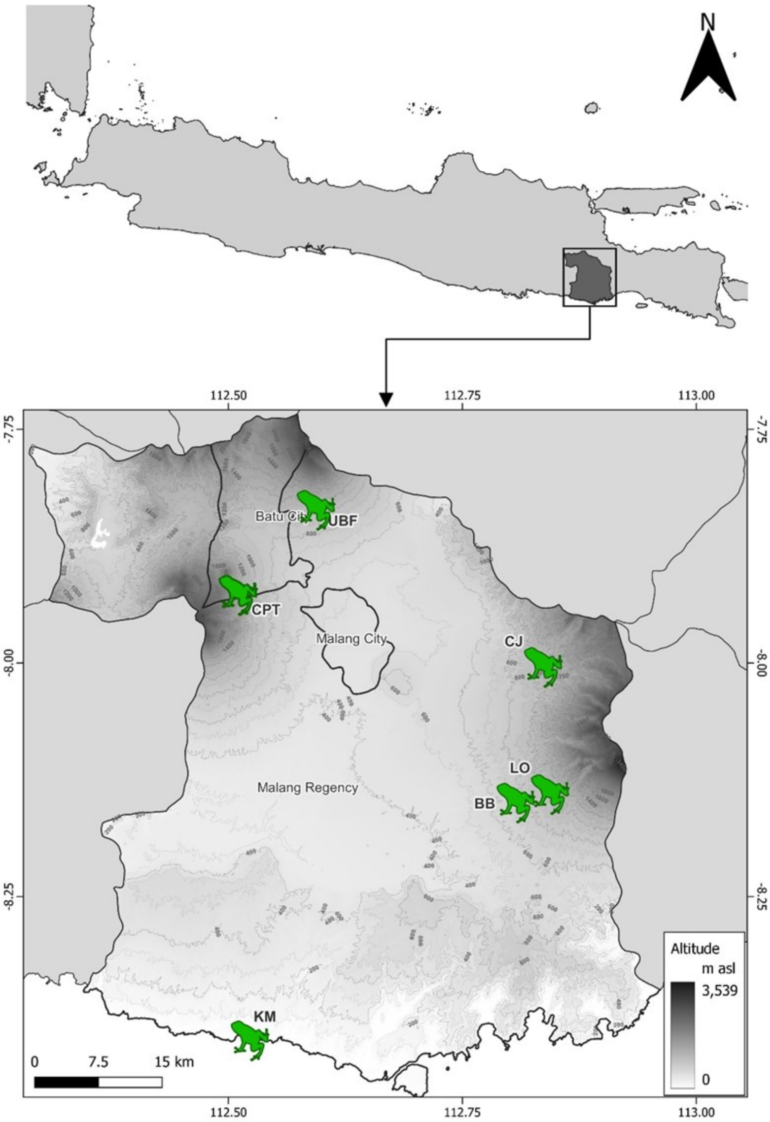

Figure 1. Field observation study area conducted in Malang Region-East Java. Note: **KM:** Kondang Merak, **BB:** Bambang village, **UBF:** Universitas Brawijaya Forest, **CPT:** Coban Parang Tejo, **CJ:** Coban Jodo, **LO**: Ledok Ombo.

The percent contribution and permutational importance of the six variables that contributed the most to the model are shown in Table 2. Based on percentage contribution, the order of the six predictor variables is as follows: tree canopy cover (46.5%), elevation (38%), precipitation of warmest quarter (13.5%), precipitation of coldest quarter (0.8%), temperature seasonality (0.7%), and slope (0.6%). In the species distribution modeling, tree canopy cover, elevation, and precipitation of the warmest quarter were found to be the most significant predictor variables

Table 2. Variable contribution (in %) of the leading six predictor variables in species distribution modelling of *R. reinwardtii* in Java.

Code	Predictor variables	Percent contribution	Permutation importance
trecov	Tree canopy cover	46.5	49.5
ele	Elevation	38	14.7
bio18	Precipitation of warmest quarter	13.5	26.8
bio19	Precipitation of coldest quarter	0.8	4
bio4	Temperature seasonality	0.7	4.9
slo	Slope	0.6	0

(Appendix 3). Moreover, the tree canopy cover was the predictor with the highest gain when used in isolation.

On the basis of the optimal predictor conditions that best represented the presence probability, a response curve composed of six leading predictor variables is generated (Appendix 4). The optimal detected predictor variables included tree canopy cover (exponentially increased after 45%), elevation (exponentially increased after 0 to 1500 m, remains stationary after 1500 m), and precipitation of warmest quarter (exponentially increased after 200 to 1200 mm, remain stationary after 1200 mm). Due to the high value of standard deviation, the other three predictor variables (precipitation of the coldest quarter, temperature seasonality, and slope) were difficult to interpret due to bias.

The results (Figures 2B-C) depicted that several large continuous forests with small forest corridors and interconnected forest patches, particularly in Western and Central Java, have the highest probability of presence (>0.75). Eastern Java is less suitable for *R. reinwardtii* because its habitat suitability is mostly in mountainous highlands with fragmented to medium-unconnected forest patches. A binary presence/absence map illustrated that only 27% (3500 out of 12829.7 ha) of Java Island is suitable for *R. reinwardtii* habitat. According to an IUCN-recommended method, *R. reinwardtii*'s extent of occurrence (EOO) is 6500 ha and its estimated area of occupancy (AOO) is 9200 ha. Our findings revealed that *R. reinwardtii* has a much smaller and fragmented habitat in Java.

Observation of habitat characteristics in Malang Region

In Malang Region our survey uncovered six cluster occurrences (Coban Parang Tejo, Coban Jodo, Ledok Ombo, Kondang Merak, Bambang village, and Universitas Brawijaya Forest), of which three were categorized as breeding sites—based on the presence eggs that accumulated in the foam nest or tadpole in nearby water bodies. We discovered that *R. reinwardtii* is arboreal and can be found perched in the tree canopy between 2.5 and 5.5 meters above the forest floor.

We also discovered that *R. reinwardtii* prefers dense vegetation, as it was observed perched in a variety of plant habitats, including herbs, shrubs, and trees, with broadleaf plants (e.g., *Colocasia* spp., *Musa* spp., *Coffea* spp., and *Persea americana*) being the most preferred. We found that they lived both solitary and communally. Males were approximately three times more common than females (male: female ratio approximately 3:1).

Based on the measured abiotic parameters (Table 3), the breeding pool's water temperature ranges from 20.5 to 24.6 °C, with a pH of 6.86 to 7.13, dissolved oxygen of 1.24 to 4.28 mg/L, and conductivity of 0.1 to 0.7 mS/cm, while the air temperature ranges from 18.5 to 25.3 °C and relative humidity ranges from 82 to 100%. Several species were found to be sympatric with *R. reinwardtii*, including *Polypedates leucomystax*, *Microhyla achatina*, *Hylarana chalconota*, and *Fejervarya limnocharis*.

Our observation reveals that it utilised a variety of habitat types (Table 4), with the majority of occurrences in agricultural areas at high

Table 3. Abiotic parameters measured on the six cluster occurrences of *R. reinwardtii* in Malang Region.

Parameter		KM	BB	UBF	CPT	CJ	LO
Microclimate	Temperature (°C)	25.3	18.7	19.7	18.5	23	22
	Relative Humidity (%)	96.1	100	100	93	86	82
Water	Temperature (°C)	24.6	22.8	20.5	-	-	-
	pH	6.90	7.13	6.86	-	-	-
	Dissolved Oxygen (mg/L)	4.28	1.24	2.21	-	-	-
	Conductivity (mS/cm)	0.7	0.1	0.7	-	-	-

altitudes (660–1094 m asl), such as the fields of Universitas Brawijaya forest and plantations in Bambang village, Coban Parang Tejo, Coban Jodo, and Ledok Ombo; the exception being Kondang Merak, which recorded a single occurrence in remaining Malang's lowland forest. However, some of these locations share similar characteristics, such as proximity to a forest and low or high tree cover density.

Based on our observations of the three breeding sites (Kondang Merak, Bambang village, and Universitas Brawijaya Forest), we determined that the spawning area was characterised by densely vegetated waters (Figures 3–5). It produces eggs and foam nests at the base of leaves and wraps eggs in the leaves of trees near bodies of water. The pools include permanent ponds, temporary water reservoirs in plantation areas, and puddles that form on roadsides.

DISCUSSION

Based on SDM (Figure 2C), *R. reinwardtii* may not be as widespread as previously thought (IUCN SSC Amphibian Specialist Group 2022). This frog is only found in a small portion of the remaining lowland forest in eastern Java, where habitat loss is highest. Sjörgen (1991) also recognised habitat loss and fragmentation as the important causes of post-metamorphic dispersion failure against anuran species. According to the IUCN SSC Amphibian Specialist Group (2022), it is assumed that the species has a large population, but data are lacking to support this. We argue that the presumption of a large population might be based on local-

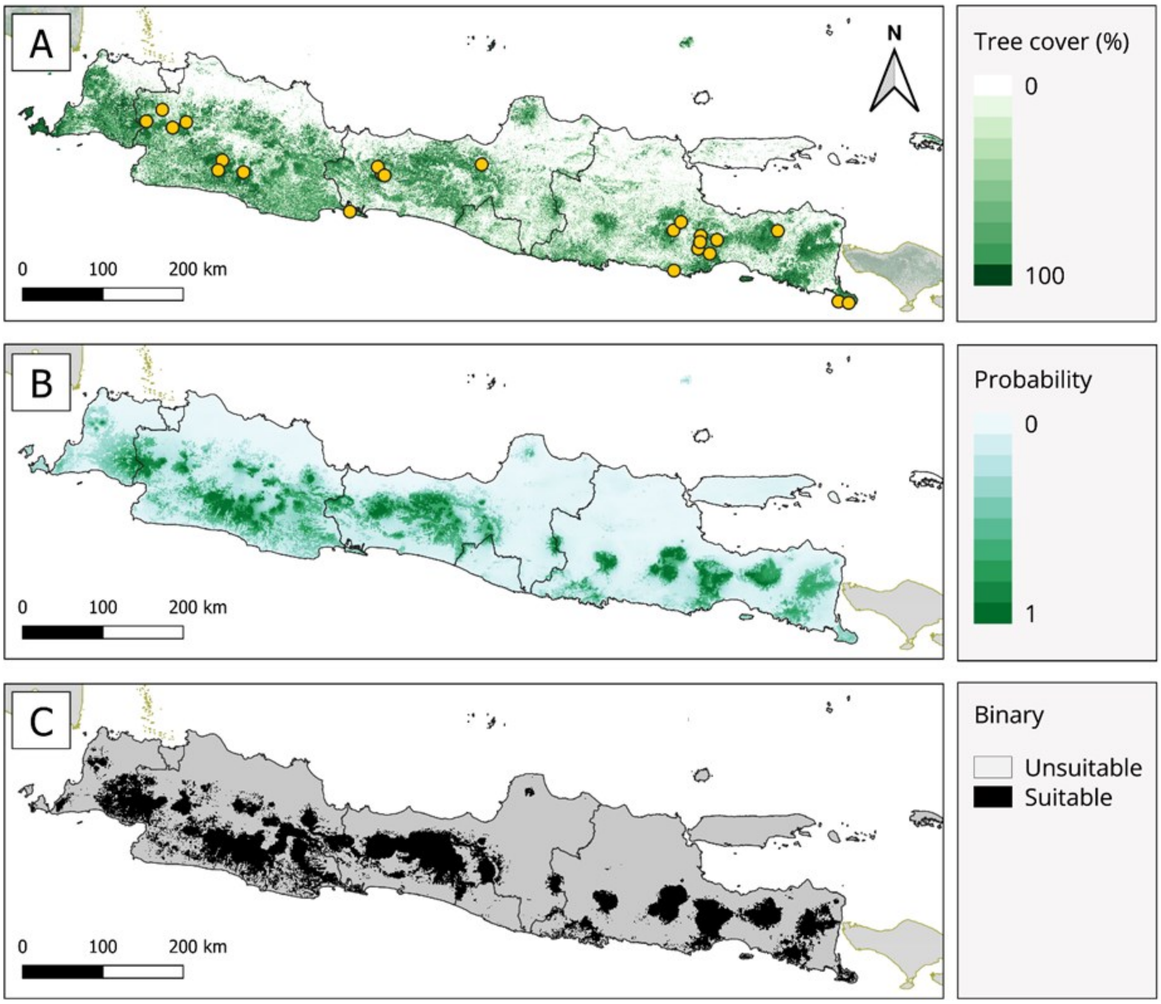

Figure 2. Geographical map showing the A) species occurrence overlayed with tree canopy cover, B) probability of presence, and C) binary habitat suitbaility of *R. reinwardtii* in Java.

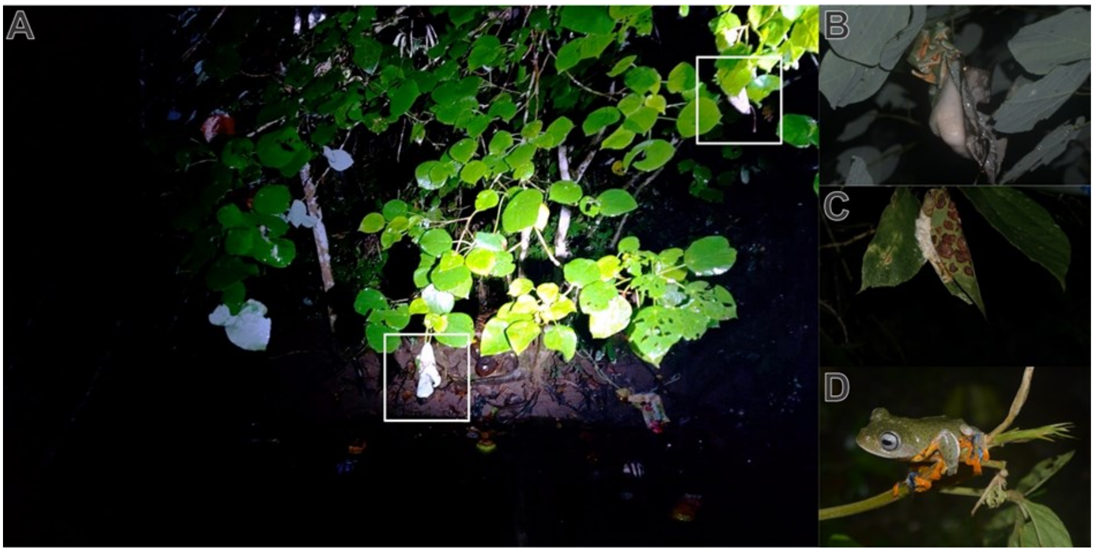

Figure 3. Observation of *R. reinwardtii* in Kondang Merak, revealing A) breeding sites in natural ponds, the squares indicate foam nests; B) male-female ovipositioning; C) remaining foam nests; and D) adult male individuals.

Figure 4. Observation of *R. reinwardtii* in Bambang village, revealing A) breeding sites in semi-permanent water storage ponds, the circle shows two male individuals perching in *Ricinnus communis*, the square shows a foam nest; B) male-female individuals in amplexus mode; C) adult male individual; and D) foam nest covered with leaves.

Figure 5. Observation of *R. reinwardtii* in Universitas Brawijaya Forest, revealing A) breeding sites in puddles that form on roadsides; B) tadpoles; C) froglets; and D) adult female individuals.

Table 4. Detailed habitat characteristics of *R. reinwardtii* observed in Malang Region.

Locality	Habitat Type	Altitude (m asl)	Breeding sites	Notes
Kondang Merak	Lowland forest	20	v	Natural ponds located near roads in primary lowland forests with dense vegetation.
Bambang	Agroforest (plantation)	660	v	Plantation areas with rainwater reservoirs around secondary high land forests with low vegetation density.
UB Forest	Agroforest (field)	1094	v	Puddles formed by vehicle paths in pine forests and coffee plantations
Coban Parang Tejo	Agroforest (plantation)	1238	-	Plantation areas around secondary highland forests with low vegetation density and close to river flows
Coban Jodo	Agroforest (plantation)	1041	-	Plantation area surrounded by secondary highland forest with low vegetation density
Ledok Ombo	Agroforest (plantation)	900	-	Plantation area surrounded by secondary highland forest with low vegetation density

ly abundant encounters, where the species descend from upper forest strata to mate and gathers, which could include up to ten to fifteen individuals during the breeding season (explosive breeder), concentrated only at those sites. Although there is no evidence that *R. reinwardtii* is experiencing a significant population decline (IUCN SSC Amphibian Specialist Group 2022), it is evident that this specialist frog relies primarily on dense canopy cover and water bodies. Consequently, it is essential to reevaluate the threat, population trend, and current distribution in light of differing viewpoints and multiple pieces of evidence.

According to numerous sources, *R. reinwardtii* is typically found at elevations ranging from 20 to 2000 m asl (IUCN SSC Amphibian Specialist Group 2022). However, the state of the forests from low-to-moderate altitude has been drastically reduced (Global Forest Watch 2021), presumably as a result of the rapid urbanization and conversion of land use (Christie 2007), leaving only the preserved highland forest and a few remaining lowland forests in the southern part of Java. This is in accordance with our habitat suitability modelling, revealing that *R. reinwardtii* is more suitable in the highlands than in the lowland forest of eastern Java (Figures 2B-C), except for western-central Java, where our prediction indicated a different result. Given the presence of intact lowland and highland forests in western Java, *R. reinwardtii* is likely to occur. In addition, Western Java has the highest proportion of vegetation and the highest density canopy; as a result, precipitation is higher and air temperatures are lower, making it an ideal habitat for *R. reinwardtii*. In comparison to eastern Java, the proportion of dense vegetation is considerably lower, and the climate is drier (PVMBG 2022). Therefore, prioritization is needed to protect Java's remaining lowland forest.

According to previous observations, *R. reinwardtii* can also hatch in vegetation near slower-flowing lotic waters (McDiarmid & Altig 1999; Kurniati 2008; Malik 2019), and it is evident from this study that *R. reinwardtii* requires artificial or natural ponds for the development of its tadpoles. Nonetheless, our observation in Malang Region, revealed an alarming threat. According to our cluster occurrence of the species breeding sites (mostly agroforestry areas), the farmers near the area frequently used agrochemicals, e.g., pesticides and herbicides, which could be harmful to the frog population (Ramadani et al. 2022). Given that a larger proportion of Java's occupants were farmers, resolving this issue could become more challenging. Therefore, an action plan and conservation strategy are necessary for the future.

We suggest that *in-situ* and *ex-situ* conservations be carried out as follows: i) establishing artificial water reservoirs at various points around natural habitats can facilitate *in-situ* conservation, particularly in agroforestry lands. The temporary reservoir could serve as a breeding site for *R. reinwardtii*, and broadleaf shade plants could be planted around the pond area to facilitate their oviposition; ii) *ex-situ* conservation—by means of reintroduction of the breed individual to the natural population, can be achieved through a captive breeding program, as in other Rhacophorid species (e.g., Tapley & Girgin 2015; Vassilieva et al. 2016; Galunder & Rodder 2018). In addition, since captive breeding programs require understanding regarding the establishment of enclosure systems (i.e.,. vivariums) and breeding protocols, it is important that we have a comprehensive understanding of a species (e.g., natural behavior, ecology, and habitat).

CONCLUSIONS

This research leads us to the following conclusions: (1) The suitable habitat for *R. reinwardtii* exists only in a small portion of fragmented habitat, which comprises only 27% of Java Island's total area. As an additional notes, (2) they inhabit areas containing or adjacent to tree cover, such as forests and agroforests in Malang Region. Nonetheless, a future action plan and conservation strategy will be required to mitigate the negative effects of a number of potential factors in these regions.

AUTHOR CONTRIBUTION

N.K., M.F., and M.A.R. were responsible for the design of the study. M.F.A. and M.F. both participated in the collection of data. M.A.R. and L.S. participated in the analysis and interpretation of the data. M.A.R. initially drafted the manuscript and N.K. supervised the entire research project as well as reviewed, edited, also proofread the final draft.

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CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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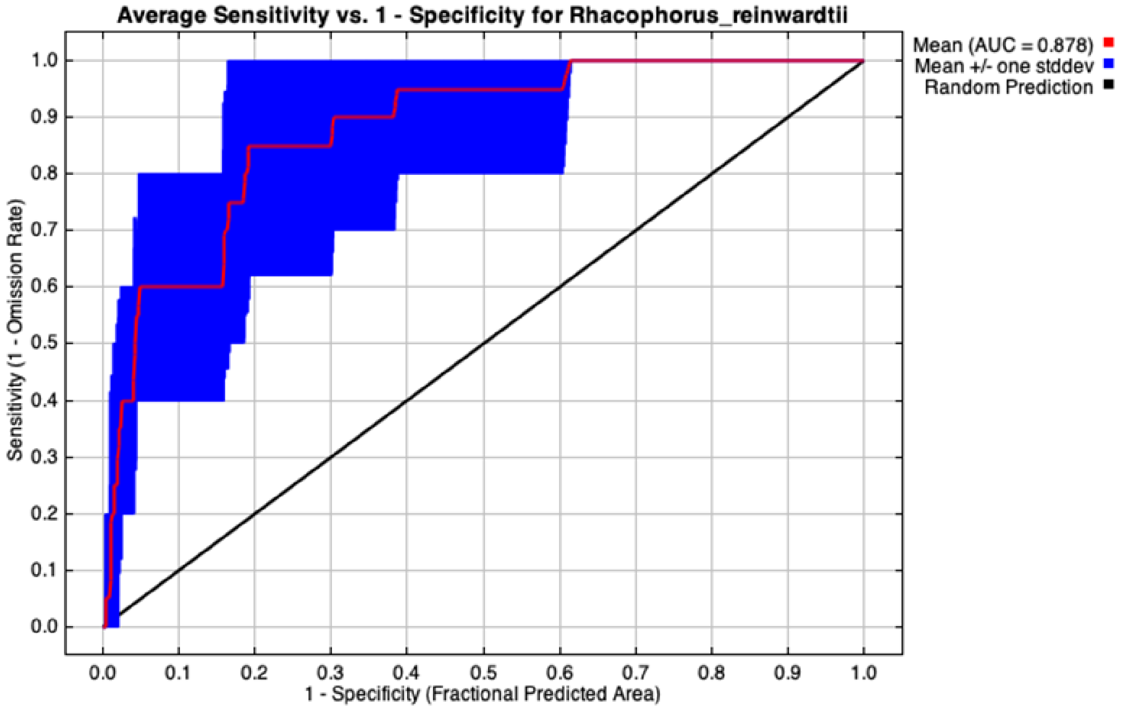
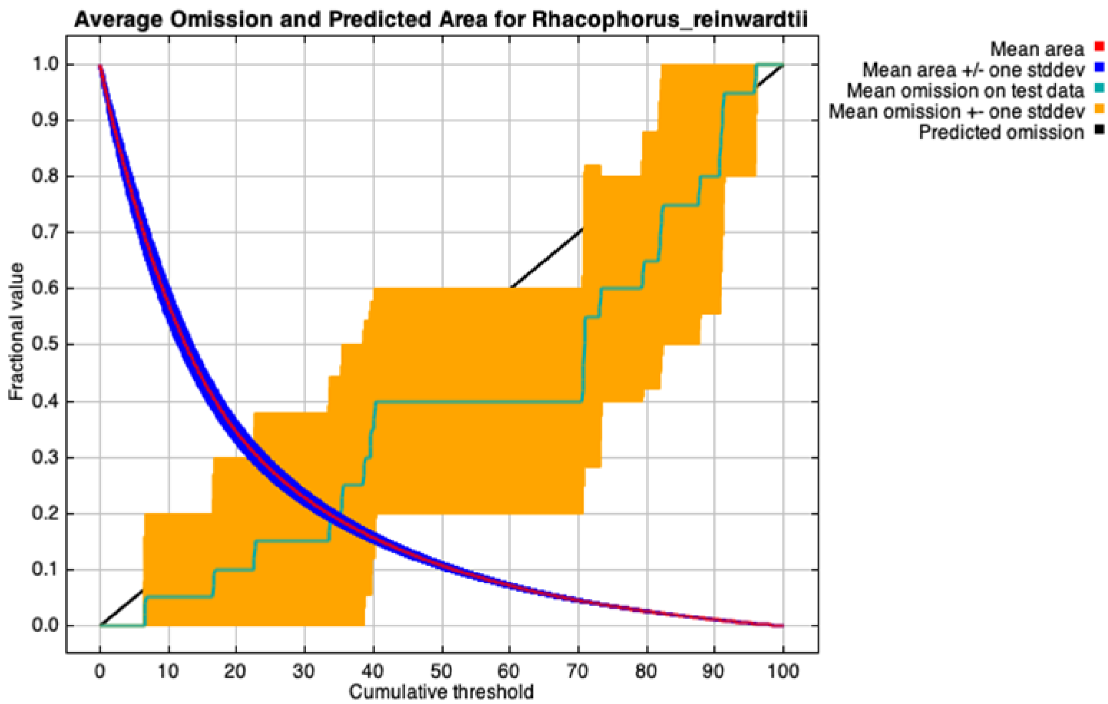
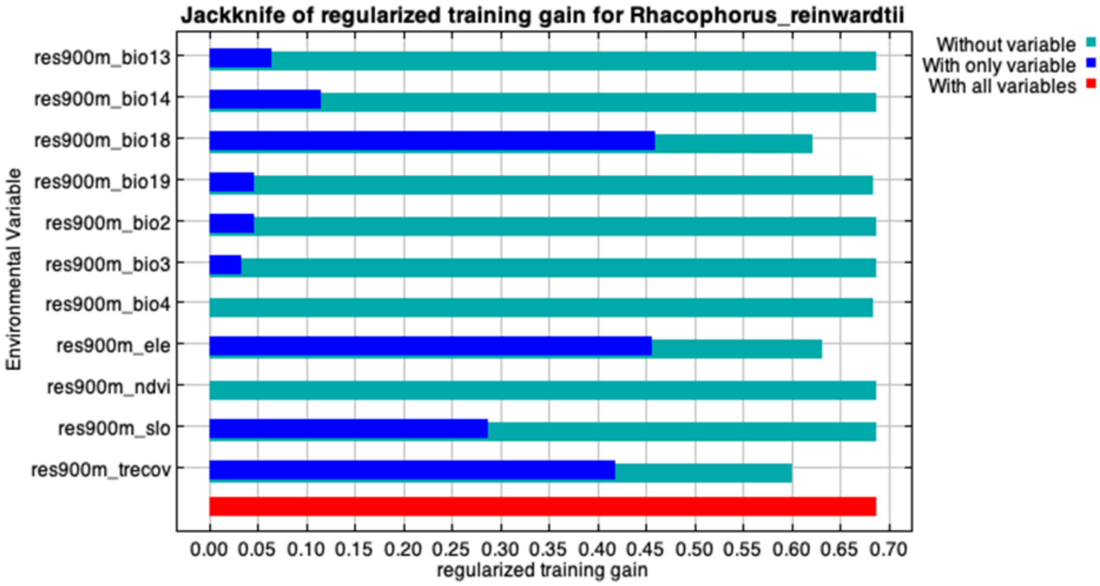
APPENDICES



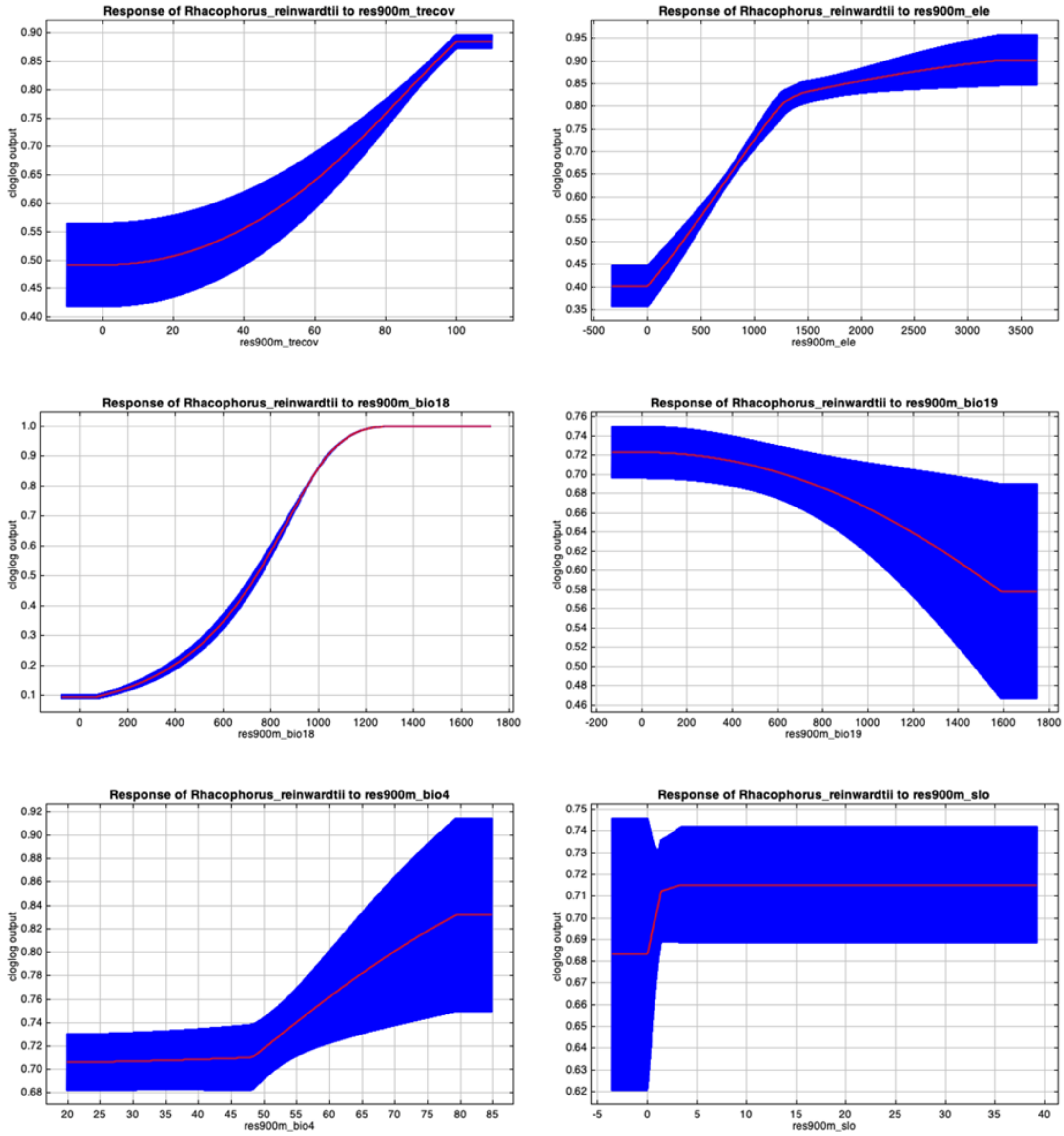
Appendix 1. Average omission and predicted area averaged for replicated runs in species distribution modelling of *R. reinwardtii* in Java.



Appendix 2. Receiver operating characteristic (ROC) curve averaged for replicated runs in species distribution modelling of *R. reinwardtii* in Java.



Appendix 3. Jacknife test of regularized training gain contribution of the predictor variables and their importance for habitat suitability in species distribution modelling of *R. reinwardtii* in Java.



Appendix 4. The response curve of the leading six predictor variables used in species distribution modelling of *R. reinwardtii* in Java.