

Filling the ecological data gap: A detailed habitat study of the neglected bleeding toad (*Leptophryne cruentata*)

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Abstract. Tohir RK, Kusrini MD, Mardiasuti A, Darmawan B, Rahman DA. 2025. Filling the ecological data gap: A detailed habitat study of the neglected bleeding toad (*Leptophryne cruentata*). *Biodiversitas* 26: 3973-3984. Effective conservation of threatened amphibians requires detailed ecological knowledge, yet many species remain poorly studied. This research focuses on the bleeding toad (*Leptophryne cruentata*), a Critically Endangered and legally protected endemic amphibian in Java, Indonesia. Despite its conservation status, ecological information about the species is scarce. We conducted habitat survey across 11 sites within Gunung Gede Pangrango National Park, West Java, from March 2023 to February 2024. A 150 m transect was randomly placed in previously known microhabitats, with environmental variables recorded twice daily and nightly (07:00-09:00) to assess diel habitat variation. We assessed elevational distribution, microclimatic parameters, hydromorphological variables, and aquatic microhabitat types. The species was observed between 1,281 and 1,892 m asl, with additional records extending to 2,089 m asl. Significant diurnal variations in air and water temperatures were found. Daytime air temperatures ranged from 15.7-21.8°C and water temperatures from 15.6-22.3°C, both significantly higher than nighttime values ($p < 0.001$). Humidity remained consistently high (75-100%) with minimal diurnal variation. Bleeding toad was associated with moderately to highly shaded habitats and diverse aquatic environments, including waterfalls, streams, swamps, and headwall seeps. A novel observation was its presence in a terrestrial habitat 260 m from the nearest stream. These findings indicate ecological plasticity and highlight critical habitat parameters essential for conservation planning. Understanding the species' elevational limits, microhabitat use, and environmental sensitivity provides a valuable foundation for habitat management, reintroduction strategies, and climate resilience planning for this imperiled montane amphibian.

Keywords: Amphibians microhabitat, ecological data gaps, headwall seeps, *kodok merah*, terrestrial

INTRODUCTION

Effective conservation of threatened species depends on understanding their distribution, threats, and the ecological factors that influence their survival, reproduction, and development (Scheele et al. 2018). However, multiple layers of bias, uncertainty, and informational gaps challenge the conservation of data-poor species (Woinarski et al. 2021). There are at least eleven key dimensions of knowledge shortfalls and biases, including a general lack of biological and ecological information (Woinarski et al. 2021). The absence of such data for numerous species poses a significant barrier to effective conservation efforts. For instance, successful captive breeding and reintroduction programs require a strong understanding of species biology and ecology, including their habitat characteristics (Stadtman and Seddon 2020; Brown et al. 2021; Bussolini et al. 2023).

Indonesia is one of the top ten countries in the world in terms of amphibian species richness (Rewild et al. 2023). Yet, it also holds the highest number of amphibian species

classified as Data Deficient (DD) (218 species) (Rewild et al. 2023). Several studies have highlighted that species classified as DD on the International Union for Conservation of Nature (IUCN) Red List often fail to reflect their actual extinction risks, with some predictions estimating that over 50% of DD species may be threatened with extinction (Borgelt et al. 2022; Edgar 2025). Therefore, addressing the lack of information on understudied species is essential and should be regarded as a matter of serious concern.

Efforts to mitigate the risks faced by data-poor species should be given greater attention. Although data may be limited, species like the bleeding toad (*Leptophryne cruentata*) have already been assigned a Red List category as Critically Endangered, based on available information such as its Extent of Occurrence (EOO) and Area of Occupancy (AOO) (IUCN SSC Amphibian Specialist Group 2019). However, it is important to recognize that this categorization often overlooks finer-scale ecological factors, such as microhabitat characteristics, which are crucial for species survival. Expanding the criteria for

assessment to include such habitat-specific details could further inform conservation efforts for this and other data-poor species. This is the case for several amphibian species in Indonesia, including *Bancet tompotika* (*Occidozyga tompotika*) and Jacobson's bubble-nest frog (*Philautus jacobsoni*) (ASEAN Centre for Biodiversity and IUCN SSC Asian Species Action Partnership 2020; Tohir et al. 2025).

Amphibian conservation in Indonesia is hindered by the limited attention and research directed toward less charismatic species, resulting in a scarcity of studies and constrained funding for conservation initiatives (Kusrini 2007a; Rahman et al. 2022). The limited development of amphibian research in Indonesia is further compounded by systemic barriers within the scientific community, ranging from bureaucratic hurdles in project planning and permit processes, to challenges in scientific collaboration and project management, alongside reduced government funding for basic research and restricted access to international scientific literature (von Rintelen et al. 2017; Tohir et al. 2025). These factors collectively hinder the internationalization of biodiversity science in the country (von Rintelen et al. 2017).

The bleeding toad is a species that merits particular attention, as it is the only amphibian protected under Indonesian law, specifically through Permen-LHK P.106/2018. Despite being legally protected and classified as Critically Endangered, ecological information on this species remains highly limited, particularly regarding its habitat characteristics (Tohir et al. 2025). The bleeding toad is threatened by habitat disturbances caused by human activities, including tourist littering at several sites such as Curug Cibeureum, Rawa Gayonggong, and Sungai Jajaway (Hasan 2022). Although the fungal infection *Batrachochytrium dendrobatidis* (Bd) has been detected at low levels in this species (Kusrini et al. 2018), it remains a concern as Bd is a major driver of global amphibian population declines (De León et al. 2019). Understanding the habitats, a species occupies is fundamental for effective management and can be crucial for ensuring their conservation (Knight et al. 2024).

This species is restricted to only known mountainous ecosystems: Gunung Gede Pangrango National Park and Gunung Halimun Salak National Park. These mountain ecosystems provide unique microhabitats essential for the bleeding toad's survival and persistence. Mountain environments are ecologically fragile systems where amphibians are especially vulnerable to synergistic threats such as climate change, land-use transformation, and emerging infectious diseases, making habitat-focused conservation research in these ecosystems critically important (Kafash et al. 2018; Haver et al. 2022; Niknaddaf et al. 2023). Due to their strong site fidelity and microhabitat specialization, amphibians are particularly dependent on specific habitat structures, which necessitates a fine scale understanding of environmental variables to inform effective conservation planning (Byrne and Silla 2023; Davis et al. 2023; Keith et al. 2025). This study aimed to address critical knowledge gaps by conducting field-based surveys focused on the bleeding toad.

Specifically, it aimed to identify the range of habitat types occupied by the species, assess its elevational distribution as an indicator of climatic sensitivity, and characterize the physical and ecological attributes of aquatic habitats associated with its presence. The findings are expected to provide valuable ecological insights that can inform national amphibian conservation policies and guide site-specific management strategies for this imperiled species.

MATERIALS AND METHODS

Study area

The research was conducted in Gunung Gede Pangrango National Park, West Java, Indonesia (Figure 1), across 11 sites: Rawa Denok (RDN), Jembatan Rawa Gayonggong 2 (JRG), Curug Cibeureum 3 (CC3), Aliran Cibeureum 1 (AC1), Curug Rasta (CRT), Curug Cibeureum Selabintana (CCS), Sungai Cimisblung (SCM), Curug Pariuk (CPR), Sungai Jajaway (SJJ), Sungai Kuta (SKT), and Curug Ceret (CCR). Site selection was based on previous records of the bleeding toad sightings obtained from earlier studies and reports from the National Park Office (Tohir et al. 2025). This study represents the most extensive site-based survey ever conducted for the bleeding toad in Gunung Gede Pangrango National Park.

Data collection

We used a multidimensional approach to determine the physical, climatic, and hydrological factors influencing the presence of the bleeding toad. We collected data on nine site-level environmental variables from March 2023 to February 2024. At each of the 11 sites, we established a 150 m transect placed randomly, based on prior detection of bleeding toad presence during preliminary surveys. The transects were located in microhabitats where the species had previously been encountered. Along each transect, environmental variables were recorded twice for each time period, two replicates during the day (07:00-09:00 AM) and two at night (07:00-09:00 PM) to capture diel variation in habitat use. The sampling effort was designed to balance spatial coverage with logistical constraints in mountainous terrain.

The 150 m transects ensured representative coverage of microhabitat variation within each site, while the diel sampling (two replicates per time period) captured essential day-night environmental shifts relevant to amphibian activity patterns. Habitat variables were defined as follows: canopy cover (percentage of overhead vegetation closure), air and water temperature (°C), humidity (%), stream width (m), depth (cm), flow velocity (m/s), and substrate composition (visual estimates of sediment types and sizes). Measurements were taken randomly whenever distinct environmental features were encountered, with a minimum of five sampling points per transect. A handheld Garmin GPS device was used to record elevation. To capture microclimatic fluctuations, we measured diurnal variation in air and water conditions.

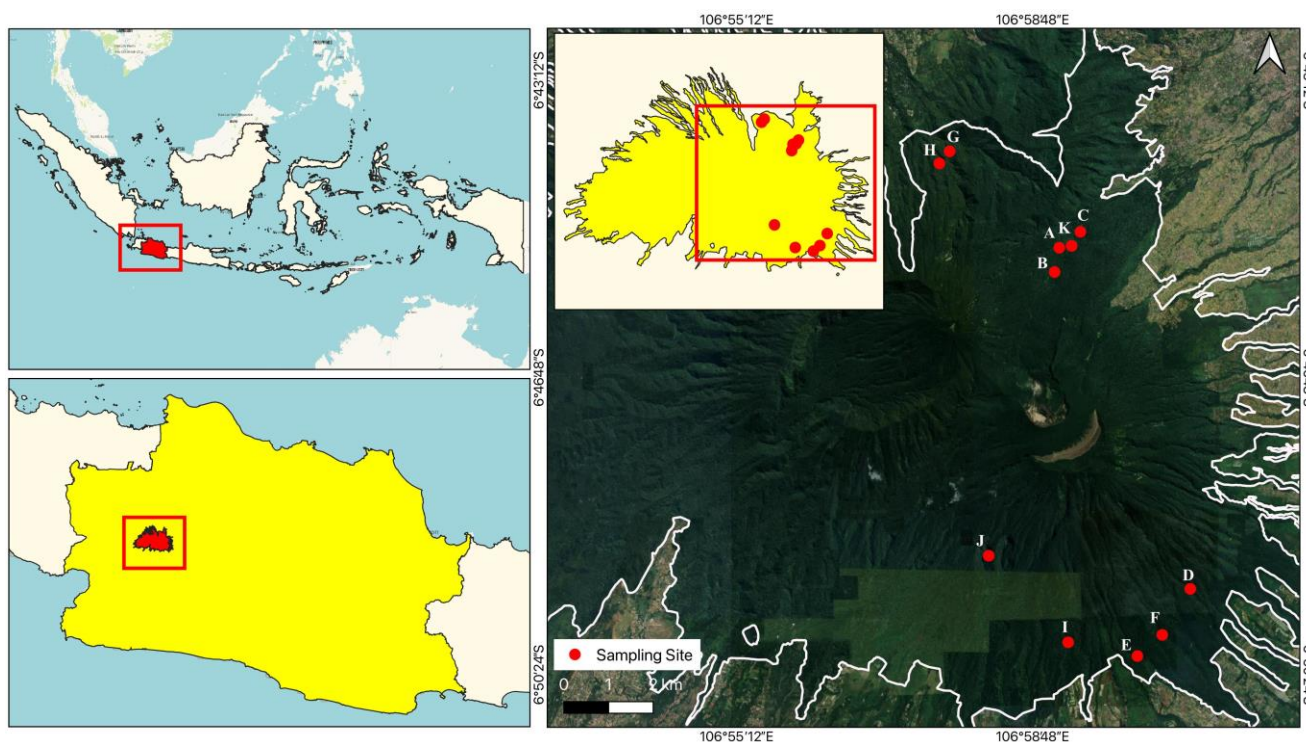


Figure 1. Geographical distribution of the 11 surveyed sites in Gunung Gede Pangrango National Park, West Java, Indonesia. A. Curug Cibereum 3 (CC3), B. Rawa Denok (RDN), C. Jembatan Rawa Gayonggong 2 (JRG), D. Curug Ceret (CCR), E. Sungai Jajaway (SJJ), F. Sungai Kuta (SKT), G. Curug Pariuk (CPR), H. Sungai Cimisblung (SCM), I. Curug Rasta (CRT), J. Curug Cibereum Selabintana (CCS), K. Aliran Cibereum 1 (AC1)

Air temperature ($^{\circ}\text{C}$) and relative humidity (%) were recorded using a dry-wet thermometer, while water temperature was measured using an infrared (gun) thermometer. Canopy cover was also assessed at each survey site to explore the relationship between vegetation structure and habitat conditions. Measurements were taken using a spherical densiometer (Robbert E. Lemmon, Forest Densiometer, Forestry Suppliers Inc), which was held upward while the observer faced four cardinal directions (north, south, west, and east). Canopy density was calculated by counting the filled squares in the densiometer's grid. A higher number of filled squares indicated denser canopy cover (Lemmon 1957).

Aquatic habitat morphology and other hydrological aspects, such as stream width, depth, flow velocity, and substrate type, were analyzed at each site. Stream width was measured using a measuring tape, while depth was determined using a calibrated rod at several transect points. Flow velocity was calculated using the float method or a flow meter. Streambed substrates were visually classified into categories such as bedrock, boulders, cobble, gravel, sand, or silt, and their relative proportions were documented. Specifically, we visually estimated the percentage composition of substrates as follows: bedrock, boulder (>256 mm diameter), cobble (65–255 mm), pebble (2–64 mm), sand (0.125–1 mm), and silt (<0.125 mm) (Huck et al. 2024). Assessing aquatic habitat morphology and stream connectivity is essential for understanding amphibian occurrence and abundance (Hamer et al. 2023).

Data analysis

We conducted descriptive and statistical analyses using RStudio (RStudio Team 2020). Prior to analysis, all data were assessed for normality and homogeneity of variance using the Shapiro-Wilk and Levene's tests, respectively. For each of the nine environmental variables, we calculated the mean, 95% Confidence Interval (CI), median, Standard Deviation (SD), and range across all eleven sites. These variables included elevation, air temperature, relative humidity, water temperature, canopy cover, stream width, stream depth, flow velocity, and substrate composition. Diurnal differences (day vs. night) in air and water temperature, and relative humidity were also summarized to assess microclimatic variation. To test for differences in environmental and hydrological parameters among spatial or temporal, we used a non-parametric test using Wilcoxon Signed-Rank Test and for the correlation test, we used the Spearman test (Holtmann et al. 2017).

RESULTS AND DISCUSSION

Physical, climatic, and hydrological features of occupied habitat

Elevational range of occupied habitats

Since the beginning of the Quaternary period (the era of modern life), mountains have served as refugia for species in response to climate change and habitat degradation driven by human population growth (Tang et al. 2018;

Ebrahimi et al. 2022). Elevation plays a key role in regulating temperature and precipitation in these areas, making it essential to closely monitor temporal changes in elevation-related variables for understanding their ecological implications (Pepin et al. 2022). The bleeding toad is a montane species highly restricted by physical limiting factors such as elevation, and is endemic to only two mountain ranges on the island of Java: Gunung Gede Pangrango National Park (GGPNP) and Gunung Halimun Salak National Park (GHSNP) (van Kampen 1923; Liem 1971; Iskandar 1998; Kurniati 2003, 2006; Kusri 2007b, et al. 2017, et al. 2021; Ningsih et al. 2013; Siregar 2016; Kusri et al. 2018; Saputro et al. 2019; Permana et al. 2020; Setiawan et al. 2021; Noer et al. 2023; Tohir et al. 2025). Reports of bleeding toad occurrences in GGPNP across various elevations have accumulated from 1923 to 2024, continuously contributing valuable updates to our understanding of the species' distribution.

In the early stages of its discovery, the bleeding toad was recorded at elevations ranging from 1,370 to 2,500 m asl (Tohir et al. 2025) However, subsequent surveys by Kusri et al. (2017) reported the species was no longer found at 2,500 m asl, suggesting a possible contraction of its upper elevation limit. More recent studies indicated that the species now occurs within an elevation range of 1,300 to 2,000 m asl (Hasan 2022; Tohir et al. 2025), reinforcing the idea of a narrowing vertical distribution. The present study further refines this pattern by providing elevation data from 11 observation sites in Gunung Gede Pangrango National Park, where the species was observed between 1,281 and 1,892 m asl (Figure 2). This is consistent with the lower elevational records noted by Kusri et al. (2017) and may point to a downward shift in the species' elevational distribution.

An observation at 2,089 m asl was recorded during fieldwork following a report from a park ranger and was subsequently validated by photographic evidence and georeferenced GPS data (Figure 6). This finding suggests the potential persistence of the species above 2,000 meters, although repeated observations are required to confirm its continued presence at such elevations. Overall, these

findings support the hypothesis of an elevational shift, particularly downward possibly in response to environmental or climatic pressures, a phenomenon that has been observed in other montane amphibians. For many vertebrates, the capacity for distributional shifts may be more constrained along elevation gradients than latitudinal ones, due to the limited time available to develop new adaptive traits (Chevin et al. 2010; Sinervo et al. 2010; Spranger et al. 2024).

This study reveals the elevational profiles of 11 current habitats occupied by the bleeding toad. The species is distributed across two elevational zones: the majority (seven locations) lie within the montane zone ($\geq 1,400$ m asl), while the remaining four are situated in the submontane zone ($< 1,400$ m asl) (Figure 2). This pattern indicates that bleeding toad tends to prefer higher elevation habitats that are cooler and more humid, consistent with the thermal and moisture preferences commonly observed among tropical montane amphibians. The site with the highest mean elevation is Rawa Denok ($1,866.8 \pm 23.57$ m asl), followed by Curug Cibereum Selabintana ($1,790 \pm 20.20$ m asl) and Curug Cibereum 3 (Cikundul) ($1,713.4 \pm 4.21$ m asl). In contrast, the site with the lowest mean elevation is Sungai Jajaway ($1,340.23 \pm 46.09$ m asl).

Locations such as Curug Ceret ($1,490.91 \pm 18.57$ m asl) and Sungai Kuta ($1,408 \pm 10.25$ m asl) represent transitional areas between montane and submontane zones. Sensitivity to temperature and humidity, both influenced by elevational gradients, may represent a key limiting factor for the distribution of this species, as noted in previous studies on tropical highland amphibians (Whitton et al. 2012). Moreover, climate change-induced shifts in vertical distribution are of particular concern for conserving highland endemic species such as the bleeding toad (Foden et al. 2013). Therefore, the long-term conservation of the bleeding toad requires special attention to montane habitats, which tend to be more stable microclimatically and less exposed to land-use change or anthropogenic disturbance. The elevation data obtained in this study may also serve as a foundation for future habitat suitability modeling under climate change scenarios.

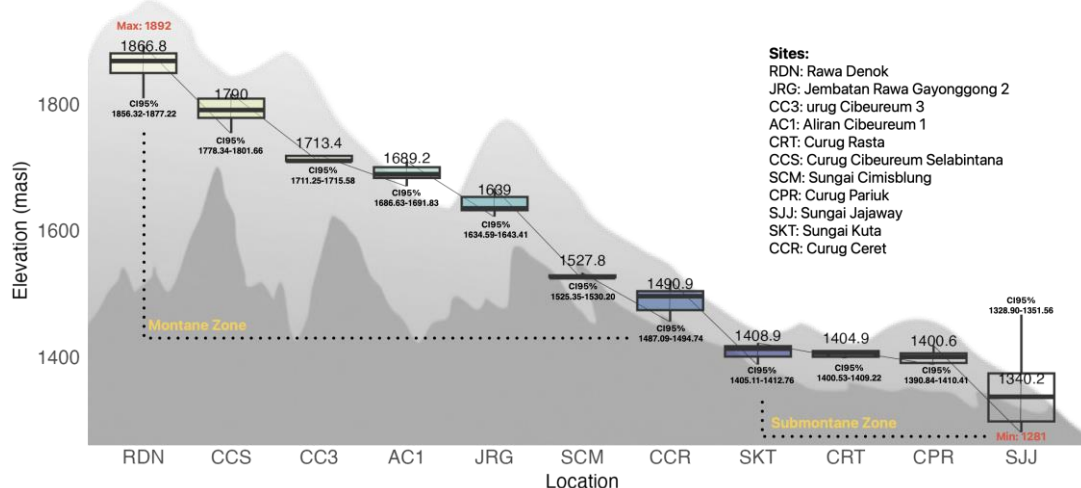


Figure 2. Elevation distribution of the bleeding toad habitat sites: Insights from 11 surveyed locations

Diurnal variation in air and water conditions across habitats

Amphibians are among the most threatened taxa under climate change (Kim et al. 2021; Pottier et al. 2025). This group is highly sensitive to thermal and hydric environmental changes due to their shell-less eggs, highly permeable skin, and a unique biphasic life cycle encompassing both aquatic and terrestrial stages (Ochoa-Ochoa et al. 2012; Duan et al. 2016). Information on the ambient temperature and humidity (both air and water) utilized by amphibian species is critically important, as it provides essential insights into the microclimatic conditions that influence amphibian physiology and activity—particularly in the context of ongoing climate change (Bartelt et al. 2022). This study presents current data on air temperature, air humidity, and water temperature (measured during both day and night) across all habitats reported to be inhabited by the bleeding toad (Figure 1). Daytime air temperatures from the 11 surveyed habitats (Figure 3.A) ranged from 15.7°C to 21.8°C, with a mean of 18.00±1.36°C and a 95% confidence interval (CI95%) of 18.24-18.78°C. Meanwhile, nighttime air temperatures (Figure 3.A) ranged from 15.4°C to 20.5°C, with a mean of 17.64±1.22°C and a CI95% of 17.40-17.88°C.

The Wilcoxon signed-rank test revealed a significant difference between daytime and nighttime air temperatures (V: 3742, $p < 0.001$). This result indicates a consistent pattern of daily temperature fluctuation, with daytime temperatures generally higher than nighttime temperatures. In addition, a Spearman correlation test was performed to assess whether air temperatures at a given site during the day were associated with those at night. This analysis showed a moderate positive correlation (ρ : 0.41, $p < 0.001$), indicating that sites with higher daytime temperatures also tended to exhibit higher nighttime temperatures. This pattern is closely related to the microclimatic characteristics of forest environments, where dense canopy cover creates more pronounced temperature fluctuations compared to open areas, thereby contributing to a buffering effect on the local microclimate (De Frenne et al. 2021). This microclimatic buffering effect helps reduce maximum air temperatures by up to 2.7°C and increase minimum air temperatures by approximately 1.2°C in temperate forests and 1.5°C in tropical forests (De Frenne et al. 2019; Santos et al. 2024). This effect is attributed to the combined influence of valley topography and canopy cover (De Frenne et al. 2021). Such a phenomenon aligns with the observed presence of the bleeding toad in river valleys characterized by moderate to dense canopy cover.

Amphibians, in some cases, prioritize humidity over temperature to regulate their body temperature and maintain hydration, as they are ectothermic animals (Mitchell and Bergmann 2016). In the case of the bleeding toad's habitat, we recorded daytime and nighttime air humidity levels across its known sites (Figure 3.B). Humidity during both periods ranged from 75% to 100%. The mean daytime humidity across all sites was 92.53±6.78%, with a 95% confidence interval (CI95%) of 91.18-93.80%. Similarly, the mean nighttime humidity was 92.55±6.89%, with a CI95% of 91.16-93.94%. The Wilcoxon

signed-rank test yielded a test statistic of V: 1547.5 with a p-value of 0.1649 ($p > 0.05$), indicating no statistically significant difference between daytime and nighttime humidity at the 5% significance level. Thus, fluctuations in humidity between day and night do not exhibit a statistically consistent pattern across the observed sites.

Spearman correlation test was conducted to assess the relationship between daytime and nighttime humidity levels. The result showed a correlation coefficient (ρ) of 0.36 with a p-value of 0.0003 ($p < 0.05$), indicating a statistically significant positive correlation. This moderate positive correlation suggests that higher daytime humidity tends to be accompanied by higher nighttime humidity, despite the absence of significant differences in their absolute values. This finding is consistent with the typical daily humidity cycle in tropical mountain regions, which is characterized by relatively low variation in atmospheric humidity (Baldysz et al. 2024). The relative stability of humidity in montane environments is attributed to local wind circulation, radiative cooling, and moisture transport through air mass movement (Fatichi et al. 2015; Chepfer et al. 2019).

Amphibians are highly sensitive to temperature fluctuations, including water temperature, which plays a critical role in growth and development during the egg and larval stages, as well as in sexual differentiation and development (Figiel 2023). We recorded water temperature usage by the bleeding toad across its known habitats (Figure 3.C) and found that daytime water temperatures ranged from 15.6°C to 22.3°C, with a mean of 18.38±1.79°C and a 95% confidence interval (CI95%) of 18.24-18.78°C. Nighttime water temperatures ranged from 15.5°C to 21.5°C, with a mean of 17.98±1.46°C and a CI95% of 17.39-17.87°C. The Wilcoxon signed-rank test yielded a test statistic of V: 1848.5 and a p-value of 0.0001277 ($p < 0.001$), indicating a statistically significant difference between daytime and nighttime water temperatures, with daytime temperatures consistently higher than nighttime.

A Spearman correlation test assessed the relationship between daytime and nighttime water temperatures. The analysis revealed a moderate positive correlation, with a Spearman's rho of 0.558 and a p-value of 1.606×10^{-7} ($p < 0.001$), indicating that higher daytime water temperatures tend to be followed by higher nighttime water temperatures. This pattern is consistent with daily fluctuations in mountain stream water temperatures. During the day, solar radiation heats the surface water, while at night, the water loses heat through radiation and convection, decreasing in temperature (Leach et al. 2023). Among the 11 study sites, only one location Rawa Denok (RDN) showed a statistically significant difference ($p < 0.05$) in both daytime and nighttime water temperatures (Figure 3.C). Rawa Denok exhibited relatively warmer water temperatures, likely due to the presence of a warm water inflow originating from an upstream source. However, it is important to note that bleeding toad abundance in RDN was the lowest among all surveyed sites, based on unpublished data from our field observations.

Hydromorphological characteristics of water bodies across habitat

Many species utilize microhabitats within montane freshwater ecosystems for various stages of their life cycles, including amphibian reproduction (Assal et al. 2021; Karami et al. 2024). Microhabitat selection and use significantly influence the survival, growth, and development of amphibians (Martins et al. 2021). Both biotic and abiotic factors, such as flow velocity, water depth, temperature, inundation duration, stream network structure, dissolved oxygen, food availability, competition, and predation, influence the abundance and distribution of amphibians (Thomas et al. 2019; Hamer et al. 2023; Ojala-Barbour et al. 2024). Understanding these factors is essential to examine the relationship between the bleeding toad and the hydromorphological conditions of its occupied microhabitats. This study characterizes key hydromorphological variables, including stream width, depth, flow velocity, and canopy cover density. The results indicate high variability in the physical characteristics of stream habitats across the 11 surveyed locations, particularly in width, depth, flow velocity, and canopy cover (Figure 4).

Stream depth at the sampling sites had a mean of 0.195 ± 0.16 m, ranging from 0.01 to 0.8 m, with a 95% confidence interval (CI95%) of 0.17-0.22 m (Figure 4.A). Stream width varied between 0.5 and 7 m, with a 95% confidence interval (CI95%) of 2.43-3.00 m. The mean stream width across all sites was 2.7 ± 1.5 m (Figure 4.B). Flow velocity in the vicinity of bleeding toad habitats ranged from 0.04 to 0.8 m/s, with a mean of 0.315 ± 0.17 m/s and a CI95% of 0.289-0.342 m/s (Figure 4.C). Rawa Denok stream exhibited the most pronounced differences compared to other sites in terms of stream width, depth, and flow velocity. This stream is characterized by its wide and deep channel with relatively fast currents. According to fluvial morphology, larger and deeper streams typically

exhibit higher flow velocities, hindering amphibian reproduction. Previous studies have identified flow velocity as the most influential factor affecting tadpole abundance in stream habitats, surpassing other variables such as substrate slope, stream depth, and water temperature (Thomas et al. 2019).

Canopy cover is a critical factor influencing amphibian abundance and community diversity, especially in riparian ecosystems (Puig-Gironès et al. 2024). Canopy cover is closely linked to the ability of amphibians to thermoregulate in relation to their environment, which in turn affects their growth, survival, and reproductive success. This information is critically important for informing conservation strategies for the bleeding toad, as canopy cover has a significant influence on amphibian body temperature and rates of water loss (Spranger et al. 2024). Canopy cover measurements at sites inhabited by the bleeding toad indicate that this species generally occupies habitats with moderate to high canopy density, ranging from 67% to 100%, with a mean of $94.4 \pm 9.29\%$ and CI95% of 92.85-95.97% (Figure 4.D).

The use of moderately to highly covered habitats may facilitate thermoregulation by allowing the toads to warm or cool their body temperature. Canopy density can have both positive and negative effects on amphibian population dynamics. While dense canopy cover may increase the risk of disease transmission, low canopy density can lead to elevated rates of dehydration (Lambrinos and Kleier 2003; Becker et al. 2012). Climate change has also been shown to influence trends in increasing canopy cover and the expansion of invasive plant species (Watling et al. 2011; Mantyka-Pringle et al. 2015; Song et al. 2018). Therefore, canopy cover can be considered an indirect factor driven by the effects of climate change, influencing amphibian population dynamics.

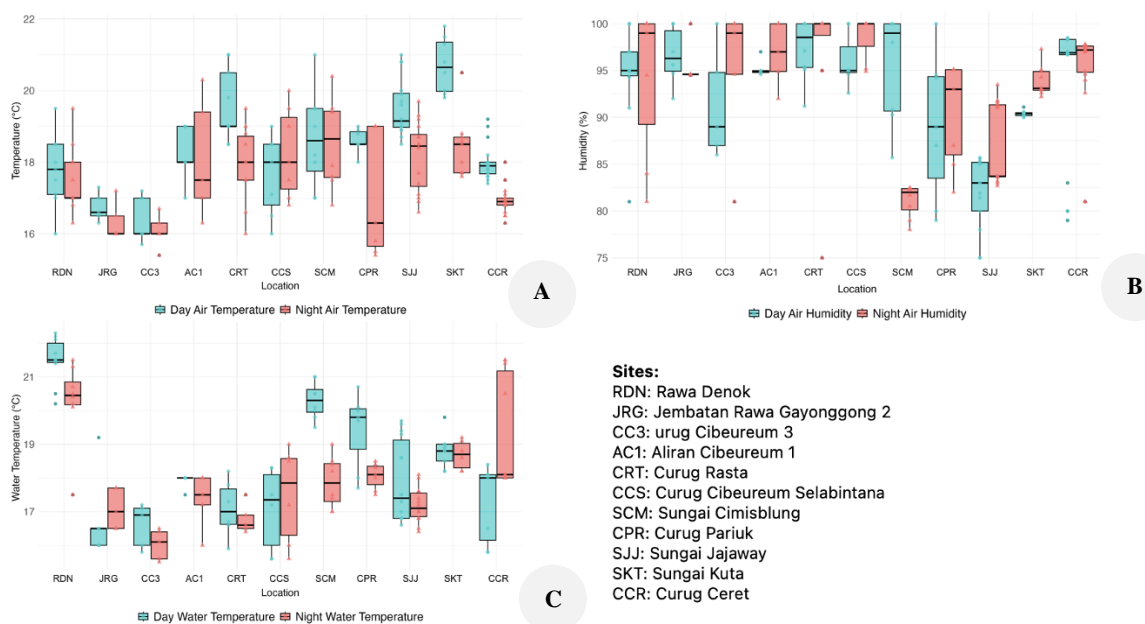


Figure 3. Diurnal variation in key microclimatic parameters across 11 habitat sites. Boxplots show daytime (blue) and nighttime (red) values for A. Air temperature (°C), B. Air humidity (%), C. Water temperature (°C)

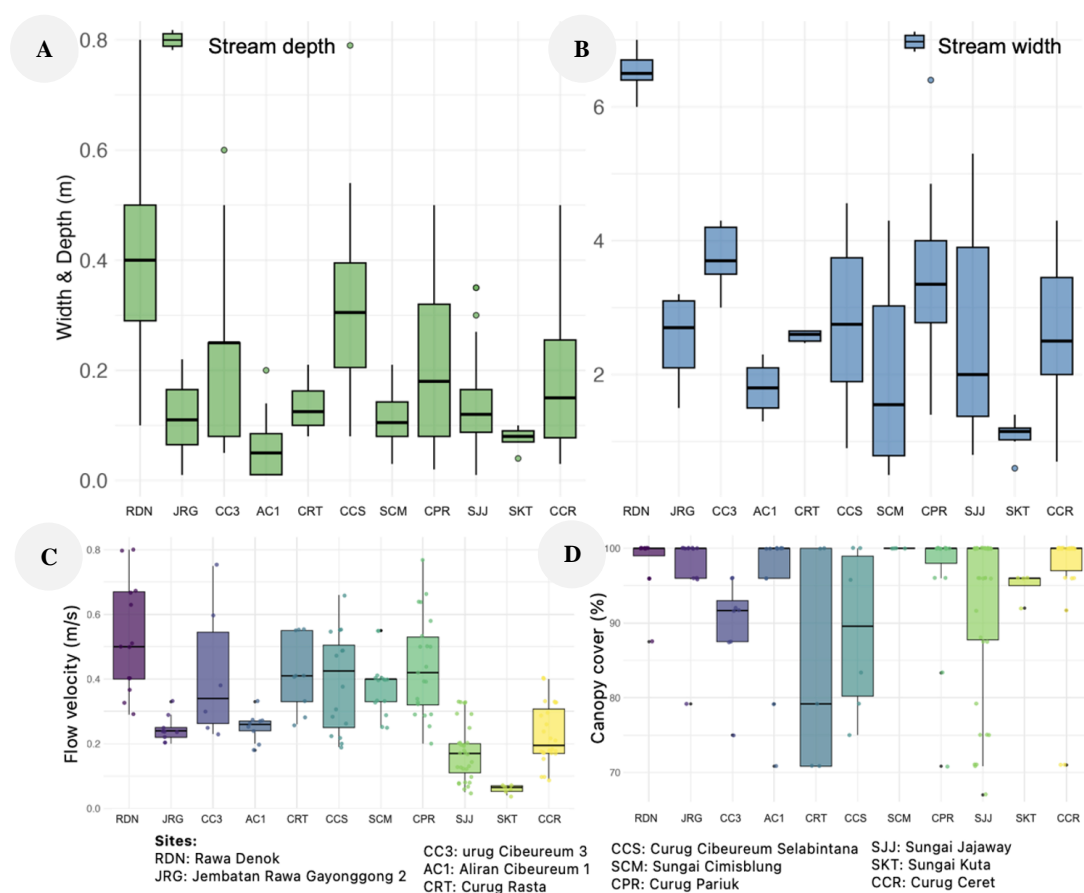


Figure 4. Variation in hydrological characteristics across 11 surveyed sites. A. Stream depth (m), B. Stream width (m), C. Stream current velocity (m/s), D. Canopy cover (%) were measured at each habitat site

Microhabitat variations across different aquatic types

Canopy cover can be considered an indirect factor driven by the effects of climate change, influencing amphibian population dynamics (Stadtman and Seddon 2020). Understanding the relationship between a species and its microhabitat is critically important, as the characteristics of aquatic environments are highly susceptible to both spatial and temporal changes driven by climate, temperature, and human activities (Bonada et al. 2006; Cecala et al. 2018). Understanding habitat selection at both spatial and temporal scales is essential for informing conservation management actions, including the design of reintroduction programs for threatened species (Brown et al. 2021).

Knowledge of the microhabitat use by the bleeding toad is critically important, particularly given its threatened status, the limited availability of ecological information, and the need to support future conservation actions such as captive breeding and reintroduction efforts (Tohir et al. 2025). This study reveals the diversity of microhabitats occupied by the bleeding. At least six types of aquatic bodies were identified as microhabitats for the species: waterfalls, large streams, medium streams, small streams, swamp/ flooded pool, and headwall seeps (Figure 5). Observations indicate that these microhabitats are utilized by the bleeding toad to support various survival strategies.

In the early records of its discovery, this species was described as inhabiting waterfalls, small streams, and riverbanks (Liem 1971; Iskandar 1998).

Based on the ecological terminology, the bleeding toad is more appropriately classified as a stream-dwelling species rather than a stream-breeding species, as it resides permanently at breeding sites and exhibits greater longitudinal movement (upstream or downstream) than lateral movement away from the stream (Hodgkison and Hero 2001). This study provides a more detailed description of the longitudinal variation in aquatic microhabitats occupied by the bleeding toad. Notably, we report the discovery of a previously undocumented microhabitat type headwall seeps associated with stream channels and waterfalls. This habitat was found on the slopes surrounding Cibereum Waterfall in Selabintana, characterized by a substrate dominated by rocks, gravel, and sand.

During the initial encounter, we heard bleeding toad calls originating from dense shrubs above a trickling water flow. Within this flow, we observed tadpoles and juveniles, leading us to infer that the site is used as a breeding habitat, supported by the presence of calling adults, larvae, and juveniles. Small hillside flows fed by springs or groundwater seepage represent an important yet often overlooked habitat for amphibians during stream-based surveys and habitat assessments (O'Donnell et al. 2007).

This observation aligns with the findings of Thurman et al. (2022), who noted that amphibians associated with headwater stream systems can be found not only in main channels, but also in isolated or hydrologically connected seeps, along riverbanks, in moss-covered margins, woody debris, and other microhabitat refugia such as sloped rocks.

Currently, at least five waterfall sites are known to be inhabited by the bleeding toad, including Curug Cibereum 3 (Cibodas), Curug Rasta, Curug Cibereum Selabintana, Curug Pariuk, and Curug Ceret (Figure 5), with waterfall heights ranging from 5 to 70 m. In these microhabitats, the toads are commonly observed on moss-covered rock walls of the waterfalls, as well as on stones or fallen logs along the pool edges. The waterfall substrates are typically composed of mossy rocks, gravel, and sand. A study on the waterfall frog (*Litoria nannotis*), which is active both during the day and at night, found that this species utilizes waterfall microhabitats for sheltering on vertical rock faces, and occasionally engages in basking behavior in well-lit areas within the spray zone (Hodgkison and Hero 2001).

This behavior is relatively similar to that of the bleeding toad in waterfall habitats, where individuals are frequently found in the spray zone.

Waterfall microhabitat appears to reduce water loss, enhance foraging opportunities and reproductive success, improve calling performance, increase egg viability, and mitigate body temperature increases associated with climate change (Mathwin et al. 2021). Further research on the daily behavior and microhabitat use of the bleeding toad in waterfall environments is needed to better understand how this species utilizes such habitats. A behavioral study of *L. nannotis* in Australia revealed that individuals use waterfall habitats for resting and basking during the day and move to vegetated areas at night to forage and seek shallow streamlets for oviposition (Hodgkison and Hero 2001). Similarly, for the bleeding toad, eggs have not been found in the splash zone or waterfall pools, but rather in shallow, slow-flowing sections of stream crevices (Hasan 2022).

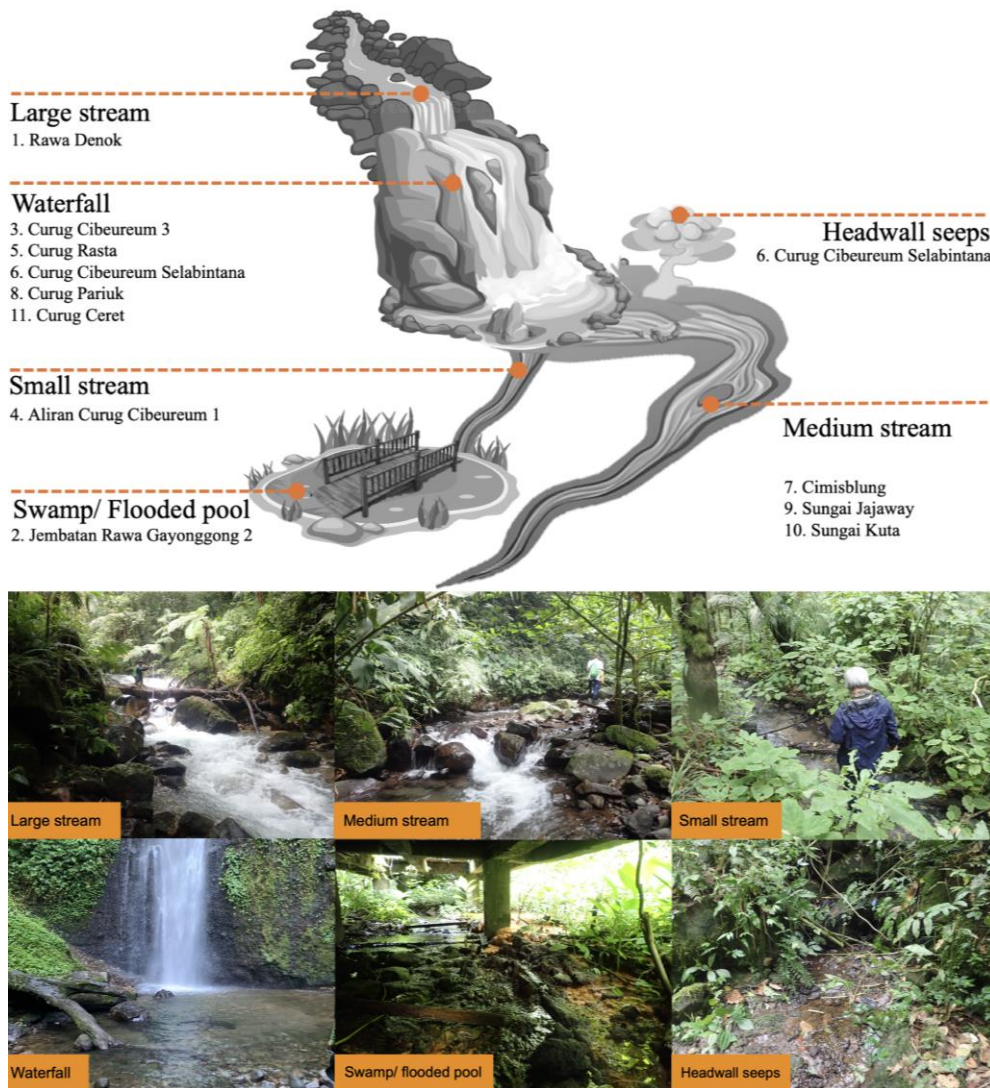


Figure 5. Illustration (top) and representative photographs (bottom) of six types of aquatic microhabitats occupied by the bleeding toad in Gunung Gede Pangrango National Park, West Java, Indonesia

The bleeding toad was also found in microhabitat formations such as swamp or floodplain pools formed by tributary streams, particularly in areas such as Jembatan Rawa Gayonggong 2 and Rawa Gayonggong 1 (Permana et al. 2020). This habitat consists of a swamp or floodplain pools with an average water depth of 0.11 m (Figure 4.A, JRG), and a substrate composed of sand and mud. The area beneath the Rawa Gayonggong 2 Bridge is a swamp fed by outflow from the Cibereum 1 waterfall tributary, characterized by its partially enclosed condition beneath the bridge structure, resulting in a relatively shaded and low-light environment. In contrast, the habitat at Rawa Gayonggong 1 was dominated by dense shrub vegetation, primarily consisting of *Etilingera elatior* (honje) (Permana et al. 2020). A relatively high number of bleeding toad individuals were recorded at this site, most of which were small in body size (indicating younger age classes), suggesting that this location may serve as a breeding habitat. Findings from the two swamp or floodplain pools sites in Gunung Gede Pangrango National Park suggest that the bleeding toad is capable of inhabiting swamp formed by stream outflows. Wetlands in montane ecosystems represent important habitats for amphibians, yet they face relatively high levels of threat due to climate change, habitat degradation, disease outbreaks, the spread of invasive fish species, and ecotourism activities (Miró et al. 2020).

River and riparian ecosystems are complex ecological systems supporting high biodiversity, particularly reptiles and amphibians (Bateman and Merritt 2020). In the context of the bleeding toad's microhabitat use, streams are crucial in connecting various habitat types, including waterfalls, swamp/floodplain pool, and headwall seeps. This study demonstrates that the bleeding toad inhabits three categories of stream habitats: large, medium, and small streams. At least five streams of varying types are still

inhabited by the bleeding toad. Large-stream habitats were observed at Rawa Denok; however, the bleeding toad did not utilize the entire stream stretch. Individuals were only found on moss-covered rocks in sheltered stream sections where the current was calm. In contrast, within medium-flow streams (e.g., Cimisblung, Jajaway, and Kuta Rivers) and small streams (e.g., Cibereum 1 tributary), the bleeding toad was more evenly distributed across the stream, occupying mossy rocks, sandy patches, and vegetated stream margins (1 m to 2 m). Lebak Saat, a stream located at 2,500 m asl, was reported to support bleeding toad populations in the 1960s. However, the stream has since dried up, and the species is no longer found there (Iskandar 1998; Kusri et al. 2017). This supports the conclusion that stream desiccation driven by climate change is a major factor contributing to declining community biodiversity (Leigh and Dady 2017).

We documented reports and evidence from national park rangers indicating the presence of the bleeding toad along a hiking trail located on a ridge, approximately 260 m away from the nearest stream (Figure 6). This new information is highly significant, as it reveals the species' ability to occupy terrestrial habitats, expanding current knowledge of its ecological flexibility. Referring to its family, Bufonidae, which is known for its broad distributional range from deserts to tropical rainforests (Pough et al. 2016). This observation warrants further investigation to understand the direction and distance of movement, the purpose of such movements (e.g., locating breeding habitats or undertaking migration), and other undocumented life history strategies. Several studies have revealed that some aquatic amphibians possess the ability to undertake migratory movements (Peer et al. 2021; Carvalho et al. 2025).

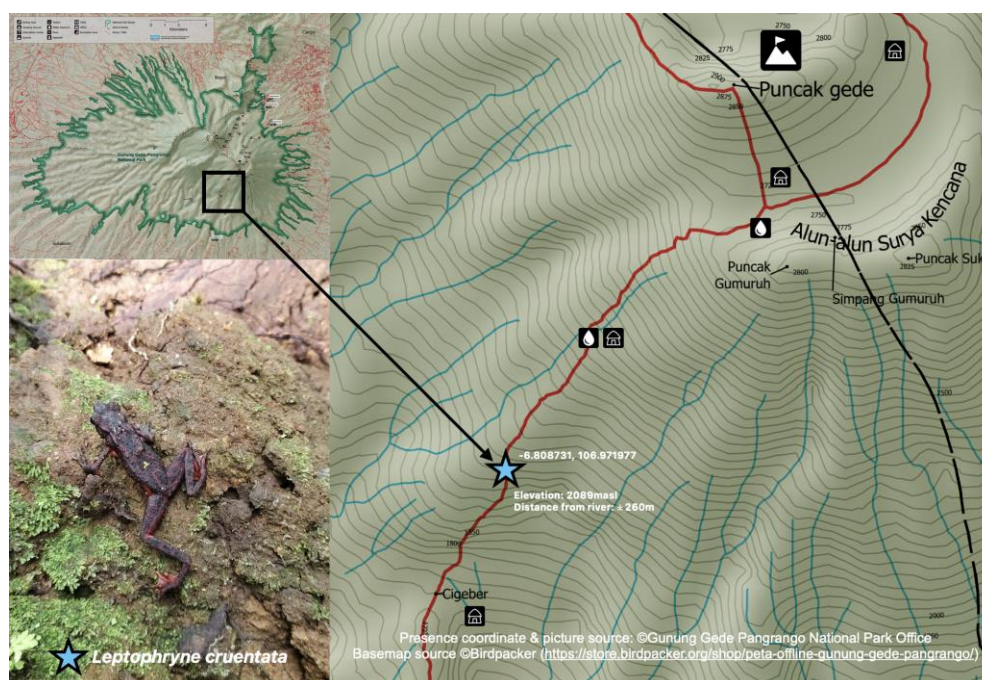


Figure 6. New report of bleeding toad distribution on the high elevation above 2000 m asl and terrestrial habitat

In conclusion, this study provides detailed ecological insights into the habitat preferences and environmental tolerances of the bleeding toad (*L. cruentata*), emphasizing its narrow elevational range and strong preference for cool, humid environments with dense canopy cover. Our findings confirm that the species is highly specific to diverse aquatic microhabitats, including waterfalls, streams, swamps, and headwall seeps. Notably, the species presence in a terrestrial habitat away from streams suggests some ecological flexibility, which could inform habitat restoration strategies.

To enhance the conservation of this species, we recommend targeted habitat restoration focused on preserving the integrity of montane stream ecosystems and riparian zones. Establishing protected areas around key habitats and monitoring these sites regularly will be crucial for ensuring long term population stability. Given the species narrow range and habitat specificity, monitoring programs should include regular assessments of temperature, humidity, and canopy cover, as well as surveys for signs of habitat degradation or encroachment. This study also highlights the urgent need for broader conservation efforts addressing Indonesia's amphibian fauna, which faces significant threats from climate change and human activity. By implementing the recommendations outlined here, we can ensure a more sustainable future for this Critically Endangered species.

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