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Poison in the nursery: mercury contamination in the tadpole-rearing sites of an Amazonian frog

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Abstract

Artisanal and small-scale gold mining (ASGM) has become a major threat for Neotropical forests. This technique for obtaining gold is a substantial driver of small-scale deforestation and the largest contributor of Hg emissions to both the atmosphere and freshwater systems globally. Previous studies have demonstrated the impacts of Hg accumulation on various aquatic ecosystems and organisms. However, its consequences in other, more discrete systems such as phytotelmata (water-holding plant structures), and the organisms therein, have so far gone unnoticed. Here, we show high concentrations of Hg (mean \pm SD: 1.43 \pm 2.19 ppm) in phytotelmata and other small pools, the aquatic microenvironments used by the Neotropical poison frog *Dendrobates tinctorius* as tadpole rearing sites. In 17 % of the cases, we detected Hg concentrations above the severe effect level (SEL = 2 ppm) for the freshwater sediments. Hg concentrations varied depending on pool characteristics and tended to increase in proximity to known ASGM sites. We did not find an effect of Hg concentration on the number of *D. tinctorius* tadpoles in a given pool. Tadpoles were found in pools with concentrations of up to 8.68 ppm, suggesting that *D. tinctorius* fathers do not avoid pools with high Hg levels for tadpole deposition. While further research is needed to determine the potential effects of Hg on tadpole development, we found an intriguing tendency for tadpoles in later developmental stages to have lower body condition when occurring in pools with higher Hg concentrations. Our findings provide evidence of relevant Hg concentrations in the terrestrial water systems used by phytotelm-breeding anurans, and highlight the need of further field and experimental studies investigating the implications of Hg contamination for tadpole development and behaviour and the overall conservation of Amazonian biodiversity.

Keywords: contaminants; phytotelmata; parental care; French Guiana; poison frog

1. Introduction

Tropical rainforests such as Amazonia are among the most biodiverse terrestrial ecosystems in the world (e.g., Grenyer et al. 2006), but also among the most threatened (Laurance et al. 2000). Many regions of the Amazon basin are characterised by naturally high levels of environmental mercury (Hg), mostly due to the geochemical characteristics of the soils and biological decay (Roulet et al. 1998; Fadini and Jardim 2001). However, anthropogenic activities such as artisanal and small-scale gold mining (ASGM; Peterson et al. 2001; Grau and Aide 2008; Hugo 2008; Kalamandeen et al. 2018) have led to dramatic increases in Hg levels in the environment over the last century (Kumar et al. 2018). ASGM is commonly illegal and, because it tends to occur in remote places, it is often in -or next to- protected areas and biodiversity hotspots (Durán et al. 2013; Cuellar et al. 2023; Palacios-Torres et al. 2018; Alvarez-Berrios and Mitchell 2019; Bax et al. 2019), making it difficult to assess timely and directly the impacts on local wildlife. Nowadays, gold mining is the single largest contributor of Hg emissions to both the atmosphere and freshwater globally (UN Environment 2019). However, other factors, such as industrial emissions, the historic application of mercury-containing pesticides and fertilisers in agriculture, and sewage irrigation, also contribute to Hg pollution (Mirlean et al., 2008; Chen et al., 2013; Bavec et al., 2014; Schneider, 2021; Fisher et al., 2023).

Hg is an environmental contaminant of global concern due to its toxicity and the risks it poses to wildlife and human health (Ashe 2012; Gibb and O'Leary 2014; Alvarez-Berrios et al. 2016; WHO 2017; Markham and Sangermano 2018). After being released to the air, land or water, and transformed by microorganisms into methylmercury (MeHg), its most toxic form, Hg can bioaccumulate in both aquatic and terrestrial organisms and biomagnify across the food webs (Kidd et al. 2011). Furthermore, gaseous Hg released in the atmosphere (for example, due to ASGM activities) can travel long distances before being deposited on surfaces and plants, or

returned to the terrestrial landscape through dry or wet deposition (Melières et al. 2003; Zhou et al. 2021; Gerson et al. 2022).

To date, assessments of Hg interactions with tropical ecosystems and biodiversity lag behind those conducted in temperate and polar areas (but see Cuellar et al. 2016; de Oliveira Drummond 2022; Palacios-Torres et al. 2018; Soe et al. 2022; as examples of studies done in the tropics). Moreover, most studies carried out to date have focused on Hg dynamics and impacts in freshwater (rivers: Webb et al. 2015; Vieira et al. 2018 and lakes: Brito et al. 2017; Gomes et al. 2020) and estuarine systems (Mol et al. 2001; Vezzone et al. 2023), while implications in other more discrete, inland aquatic systems remain unexplored. Phytotelmata, for example, are small bodies of water contained in vegetation structures (e.g. leaf axils, fallen bracts, holes in trunks and roots), which provide essential habitat for the breeding, feeding, refuge and development of a wide range of species (Varga 1928; Fincke 1999; Kitching 2000; Lehtinen et al. 2004), including many insects (Orr 1994; Kitching 2000; Campos and Fernández 2011), and amphibians (e.g., Kam et al. 1996; Schiesari et al. 2003; Lehtinen et al. 2004, Heying 2004). Because these small pools are directly formed by rainwater and accumulate decomposing leaf litter from the surrounding canopy (Kitching 2000), it could be expected that they accumulate high concentrations of Hg over time, particularly in the context of ASGM (Malm et al. 1998; Melières et al. 2003; Gerson et al. 2022). Furthermore, differences in the physico-chemical properties among phytotelmata, as well as in their morphology and durability, could influence the amount of Hg, its chemical form and, ultimately, its toxicity and absorption. For example, acidic pH conditions, warmer water temperatures, high dissolved organic carbon, and low dissolved oxygen and electrical conductivity, have been shown to increase Hg methylation (i.e. MeHg) in water and sediments in some environments (Mauro et al., 1999; Bank et al., 2007; Jardine et al., 2013; Brito et al., 2017).

Neotropical poison frogs of the family Dendrobatidae are, in most part, dependent on phytotelmata as tadpole-rearing sites (Summers and McKeon 2004; Carvajal-Castro et al. 2021) due to their elaborate parental care behaviour. Generally, one of the parents takes care of the terrestrial clutches and, upon hatching, transports tadpoles on its back to the bodies of

water, such as phytotelmata, where offspring will remain confined until metamorphosis (Weygoldt 1987; Summers and McKeon 2004). Selecting a good tadpole-rearing site is determinant for tadpole survival, and some poison frog species are known to base such decisions on chemical cues, either for detecting (and avoiding) potentially cannibalistic conspecifics or predators (Schulte et al. 2011; 2014) or for finding new deposition sites in the forest (Serrano-Rojas and Pašukonis 2021; Peignier et al. 2023). Because tadpoles are omnivores, largely feeding on detritus and larvae of insects and frogs (Summers and McKeon 2004; Grant et al. 2006; Rojas 2014), have a thin and highly permeable skin (Rowe et al. 2003; Quaranta et al. 2009), and need to process water for respiration, they can be particularly exposed to, and affected by, both waterborne and dietary contaminants (Burger and Snodgrass 2001; Unrine 2007; Rissoli et al. 2016). Thus, Hg may pose a considerable threat to tadpoles of this and other species developing in phytotelmata if adults cannot detect and avoid Hg contaminated water bodies (e.g., via chemical cues) for oviposition or tadpole deposition. Previous studies in subtropical and temperate zones have reported high concentrations of Hg bioaccumulated in amphibian larvae through environmental exposure and diet (Byrne et al. 1975; Unrine et al. 2007; Bank et al. 2007), and negative effects of Hg on development (i.e. smaller body size, delayed metamorphosis, higher prevalence of malformations) and survival (Unrine et al. 2009; Shi et al. 2018).

Here, we measured the concentrations of Hg found in different types of phytotelmata (hereafter referred to as “pools”) over six different locations in French Guiana, a French Overseas Territory with high prevalence of ASGM (Boudou et al. 2006; Grimaldi et al. 2015). We covered a gradient from pristine/protected forest to disturbed, semi-urbanised areas, at variable distances from known ASGM sites, to assess whether pools potentially used as tadpole-rearing sites contained Hg and, if so, in what concentrations. We focused on pools that could be used by the dyeing poison frog, *Dendrobates tinctorius*, as males of this species use phytotelmata with a wide range of physical and chemical properties for tadpole deposition (Fouilloux et al. 2021). *Dendrobates tinctorius* parents can also use available water-filled plastic containers (e.g., Rojas 2015) to deposit their tadpoles. Thus, we also sampled artificial

pools in this study. Because ASGM activities use Hg to facilitate gold extraction, we expected to find higher Hg concentrations in closer proximity to ASGM sites. Furthermore, because phytotelmata in tree holes, both in live and fallen trees, are generally more stable over time than other plant structures such as fallen palm bracts, they can accumulate higher amounts of rainwater and litterfall; thus, we predict this type of phytotelmata to have the highest Hg concentrations. Finally, we also recorded the presence/absence, number and size of *D. tinctorius* tadpoles in each of the pools sampled to investigate (1) potential parental avoidance of pools with high Hg concentrations, as seen in other anuran species which avoid herbicide polluted pools for oviposition (e.g. Takahasi 2007, Vonesh and Buck 2007); and (2) the potential impact of Hg on larval body condition. If parents are able to detect Hg in the water, we predict pools with higher Hg concentrations to hold fewer (or no) tadpoles than pools with low concentrations or no Hg. Moreover, if Hg negatively affects tadpole growth and development, as previously shown in some temperate species (e.g. Unrine et al. 2009), we predict pools with high Hg levels to harbour tadpoles with poorer body condition than those in pools with little or no Hg.

2. Material and methods

2.1 Study species

Dendrobates tinctorius (Figure 1a) is a rainforest dweller endemic to the Eastern Guiana Shield (French Guiana, Suriname and Northwestern Brazil), where it can be found on elevated ridges or plateaus up to 600 m above sea level (Noonan and Gaucher 2006). They are diurnal, and characterised by a highly variable bright colouration coupled with the possession of skin toxins (Rojas and Endler 2013; Lawrence and Rojas et al. 2019) and an elaborate male parental care. Their clutches are laid terrestrially, on the leaf litter or within hollow fallen logs or tree roots, and taken care of by the male until tadpoles hatch (Rojas and Pašukonis 2019). Upon hatching, males transport the tadpoles on their back to bodies of water contained in the vegetation (i.e., phytotelmata; Rojas 2014; 2015; Fouilloux et al. 2021). Once deposited, tadpoles will remain in these pools (Figure 1b-d) selected by their father until metamorphosis,

which is reached after 2-3 months (Rojas and Pašukonis 2019). *D. tinctorius* was declared a protected species in French Guiana in 2020 (Decree No. TREL2032100A from November 19th 2020).



Figure 1. A male *Dendrobates tinctorius* carrying two tadpoles on his back (a) and the diversity of sampled pool types used as tadpole-rearing sites. Pools used include phytotelmata formed in palm bracts (b), trunk/root holes (c) and fallen trees (d), as well as artificial pools formed in anthropogenic litter (e).

2.2 Study areas and field sampling

From January to March 2023, we collected a mix of water and organic matter samples from pools in six different study sites in French Guiana. These sites are known to hold *Dendrobates tinctorius* populations, and are found under different degrees and types of disturbance, and at variable distances from the closest ASGM site (see Table 1). In each study site, we searched for pools along transects between 10 and 15 km in length, targeting suitable microhabitats such as fallen trees and trees with buttresses or hole structures. When a pool was found, we

recorded its type according to the following three categories for phytotelmata: holes in fallen trees; holes in trunks and roots of living trees; and fallen palm bracts. In addition, we also included pools formed in rocky substrates and artificial plastic or metallic containers, as *D. tinctorius* tadpoles can be found in both types. Artificial pools were found in large numbers in Petit Matoury and Cacao, and only a small random subset was sampled. We obtained the distance to the closest ASGM site for each study area from the French 'Unité Spécialisée Nature', National Office for Forests (ONF).

Table 1. Overview of the study sites, the type of surrounding habitat, the distance to the closest ASGM site and the number of pools sampled.

Location	Surrounding habitat	Distance to closest ASGM site (km)	Number of sampled pools
Petit Matoury	Urban	32	13
Mont Fortuné	Urban	31	28
Cacao	Agricultural	1.6	16
Mataroni	Logging roads	16	14
Mont Favard	Natural Reserve	27	13
Camp Pararé	Natural Reserve	15	14

In addition, we noted each pool's morphological and physico-chemical characteristics, and its GPS position. Morphological parameters included height from the ground to the pool edge, largest width and length parallel to the water surface, and water depth. Based on these measurements, we calculated the surface area per water volume ratio (SA/V) of each pool

using the formula for a semi-ellipsoid. Physico-chemical parameters included water pH, alkalinity (KH), and water temperature. Water temperature and pH were recorded with a pH metre (pH20, Apera Instruments®), and KH was measured using aquarium water testing strips (JBL EasyTest).

For each pool, we collected a 4 ml mixture of water and sedimentary organic matter with a sterile syringe. Tadpoles use sedimentary organic matter for feeding and hiding, and may be therefore exposed to Hg concentrations from both water and organic matter. Further, we recorded the presence/absence of tadpoles and, when present, attempted to assess the total number of tadpoles and noted the species. If *D. tinctorius* tadpoles were found in the pool, we used a net or plastic spoons to catch five individuals representing the observed variation in developmental stage. Each captured tadpole was weighed to the nearest 0.01 g and photographed over graph paper to measure its length afterwards. We also noted each tadpole's developmental stage *sensu* Gosner (1960), and classified it into three categories (early pre-metamorphosis: 25-29; late pre-metamorphosis: 30-35; and pro-metamorphosis/metamorphic climax: 36-45) to account for its nonlinear nature. This means that larval development time to change from e.g. stage 26 to 27 is not equivalent to that from stage 42 to 43. Documenting tadpole developmental stages also allowed us to account for differences in exposure times to Hg in the pool. All tadpoles were returned to the same pool where they were caught immediately after. Capture and sample collection were done under a permit from the Direction Régionale des Territoires et de la Mer de Guyane (R03-2022-12-28-00004) after assessment by the CSRPN, the regional scientific committee. Procedures employed at Nouragues Research Station were also approved by the Reserve's scientific committee.

2.3 Mercury analyses

Total Hg (THg) was quantified in 400 µL of water and sedimentary organic matter using a Direct Mercury Analyser (DMA-80, atomic absorption spectrometer, Milestone). The method

was validated by the analysis of certified reference material (CRM) TORT-3 (Lobster hepatopancreas from the National Research Council of Canada (NRCC); certified Hg concentration 0.292 ± 0.022 ppm dw) at the beginning and the end of analytical cycle and after every 10 samples. Measured values for TORT-3 were 0.298 ± 0.007 ppm dw ($n = 15$). Blanks were included at the beginning of each analytical run and the limit of detection of the DMA-80 was 0.001 ng Hg. Mercury concentrations are presented in ppm.

2.4 Statistical analyses

We checked the normality of our data using a Shapiro-Wilks test, and log-transformed Hg concentration data to achieve normality of residuals. We used a linear mixed-effect model with a Gaussian distribution to assess the relationship between measured Hg concentration (response variable), distance to the closest known ASGM site, pool type, height, pH and SA/V (explanatory variables). We included study site as a random intercept to account for unmeasured differences between locations. We excluded KH and water temperature from our analysis since its variation between pools was very low and independent from the type of pool and the study site (see Table S1).

To investigate the effect of Hg concentration on the probability of presence of *D. tinctorius* tadpoles and tadpoles from all species recorded, we used generalised linear mixed-effects models (GLMM) with a binomial error distribution. In addition to Hg concentration, we included the pool type and pool traits (i.e. SA/V, height and pH) as fixed effects, and study site as random effect. For the analysis with only *D. tinctorius*, we also included the number of tadpoles present in the water body as a fixed effect, as it may influence parental care decisions (Rojas 2014). Further, we studied the effect of Hg on the total number of *D. tinctorius* tadpoles per pool using a GLMM with a Poisson distribution, including Hg concentration, pool type and pool traits as fixed effects, and study site as random effect. We repeated this last analysis for all tadpole species together.

We calculated the body condition of tadpoles using the scaled mass index (Peig and Green 2009, 2010). For that, we used tadpole body length measured from the tip of the snout to the most posterior point of the body, excluding their tail to avoid errors due to tail injury. To analyse the effect of Hg concentration on tadpole body condition, we used a linear mixed-effects model with a Gaussian distribution. We included Hg concentration, number of tadpoles in the pool and pool traits as fixed effects, and pool ID nested in study site as a random effect to control for non-independence of the data. In addition, because body mass and SVL vary with Gosner stage as tadpoles approach metamorphic climax, we included stage categories and the interaction with Hg in our model to control for possible differences in body condition due to developmental stage rather than Hg levels.

For all analyses, we used the variance inflation factor to identify multicollinearity between independent variables in the different models, and removed correlated variables when required ($VIF > 3$; Zuur et al., 2010). We used model selection based on Akaike's Information Criterion for small sample sizes (AICc) (Burnham et al. 2011), using the R package 'MuMIn' (Barton 2020), selecting the model with the lowest AICc value. If two or more of the highest ranking models were within $\Delta AIC < 4$, we performed model averaging (Symonds and Moussalli 2011). Parameters that included zero within their 95% CI were considered uninformative (Arnold 2010). We accepted statistical significance at $p < 0.05$ and trends at $p < 0.1$. All statistical analyses were performed using the software R version 4.1.2 (R development Core Team 2022).

3. Results and discussion

Total Hg levels found in the water and sediments of all pools in this study (including rock and artificial pools) were relatively high, ranging from 0.007 to 13.040 ppm (mean \pm SD: 1.43 ± 2.19 ppm), being highest in Cacao and lowest in Mont Fortuné (Table 2). The concentrations found in this study are comparable to those reported in polluted water bodies near mining sites (Jiang et al. 2006; Soe et al. 2022) or abandoned chlor-alkali plants (Degetto et al. 1997; Song et al. 2018) elsewhere. One sample was under the detection limit. An outlier value of 172.06

ppm belonging to an artificial pool (a rusty paint barrel) was removed from the main analysis, as we were unable to determine whether this high value could be attributed to a measurement error or to leftovers of the previous content therein. In 17% of our samples, Hg concentrations were above the severe effect level (SEL = 2 ppm) for freshwater sediments reported by the US National Oceanographic and Atmospheric Administration (Buchman 2008). Above this level, sediments are considered heavily polluted and adverse effects are expected to occur in the majority of sediment-dwelling organisms.

Table 2. Number of pool types sampled by study site, mean and standard deviation of Hg concentrations (ppm), total number of *D. tinctorius* tadpoles observed and number of *D. tinctorius* tadpoles sampled per study site. Columns coloured in green correspond to phytotelmata.

Study site	Type of pool					Mean \pm SD Hg concentration (ppm)	Number of <i>D. tinctorius</i> tadpoles observed	Number of <i>D. tinctorius</i> tadpoles sampled
	Palm bract	Tree hole/ roots	Fallen tree	Rock hole	Artificial			
Petit Matoury	2	4	0	0	6	1.38 \pm 2.42	147	11
Mt Fortuné	0	25	3	0	0	0.77 \pm 0.87	30	14
Cacao	0	6	5	5	2	2.72 \pm 3.96	3	2
Mataroni	0	7	6	0	0	1.70 \pm 1.94	15	11
Mt Favard	7	4	0	1	0	1.30 \pm 1.47	16	8
Camp Pararé	2	12	1	0	0	1.08 \pm 0.87	16	9

Pools in locations closer to known ASGM sites tended to accumulate higher Hg concentrations (Table 3, Figure 2a). Among the pools sampled, those in Cacao, which is only 1.6 km from the closest known ASGM site, had the highest concentrations of Hg. All other locations were ≥ 15 km from the nearest known ASGM. These findings suggest a potential link between ASGM activities and Hg pollution in pools. Another possible explanation for the high Hg concentrations found in Cacao could be the (historic) use of Hg-containing fungicides or phosphorus fertilisers with Hg impurities in the agricultural fields that dominate the landscape (Mortvedt 1995; Turull et al. 2018; Tang et al. 2018). Although Hg-containing fungicides have been banned in the EU since 2017 (<https://minamataconvention.org>), they were commonly used during the second half of the 20th century and can remain in the soil for long periods of time (Murphy and Aucott 1999; Horowitz et al. 2014; Ali et al. 2019; Schneider 2021). Pools in Mont Fortuné, in contrast, had the lowest values of Hg. This could be explained by the fact that, despite being surrounded by urban areas, this location is not directly adjacent to agricultural fields or close to illegal ASGM activities, which often take place in remote locations (Durán et al. 2013; Rahm et al. 2014).

Deeper pools in relation to their surface area, as well as pools closer to the ground, had higher concentrations of Hg (Table 3, Figure 2b-c). Pools in rock holes (mean \pm SD: 3.38 ± 4.76 ppm) and artificial pools (2.32 ± 2.92 ppm) generally showed the highest levels of Hg, followed by fallen trees (1.59 ± 2.21 ppm) and tree holes (1.20 ± 1.83 ppm). Palm bracts had the lowest Hg concentrations (0.55 ± 0.50 ppm) (Figure 2d). After accounting for pool height and SAV, pool type and pH were uninformative in explaining variation in Hg concentration (Table 3). However, pool type and pool characteristics (SAV and pool height) were interrelated (Figure S1, S2), and it is possible that other pool characteristics also influenced Hg concentrations (e.g., durability, material of artificial pools) (Fouilloux et al. 2021). For example, shallow pools in relation to their volume (i.e. high SAV), which had the lowest Hg concentrations, were mostly represented by palm bracts (Figure S1). Palm bracts are typically ephemeral and may dry out and decompose rapidly (Fouilloux et al. 2021), having relatively little time to

accumulate Hg through rain and leaf litter. In contrast, water pools formed in rock holes may have naturally higher levels of Hg due to both, their permanent nature when compared to degrading plant structures, and their direct contact with Hg-rich soils (Roulet et al. 1998; Fadini and Jardim 2001).

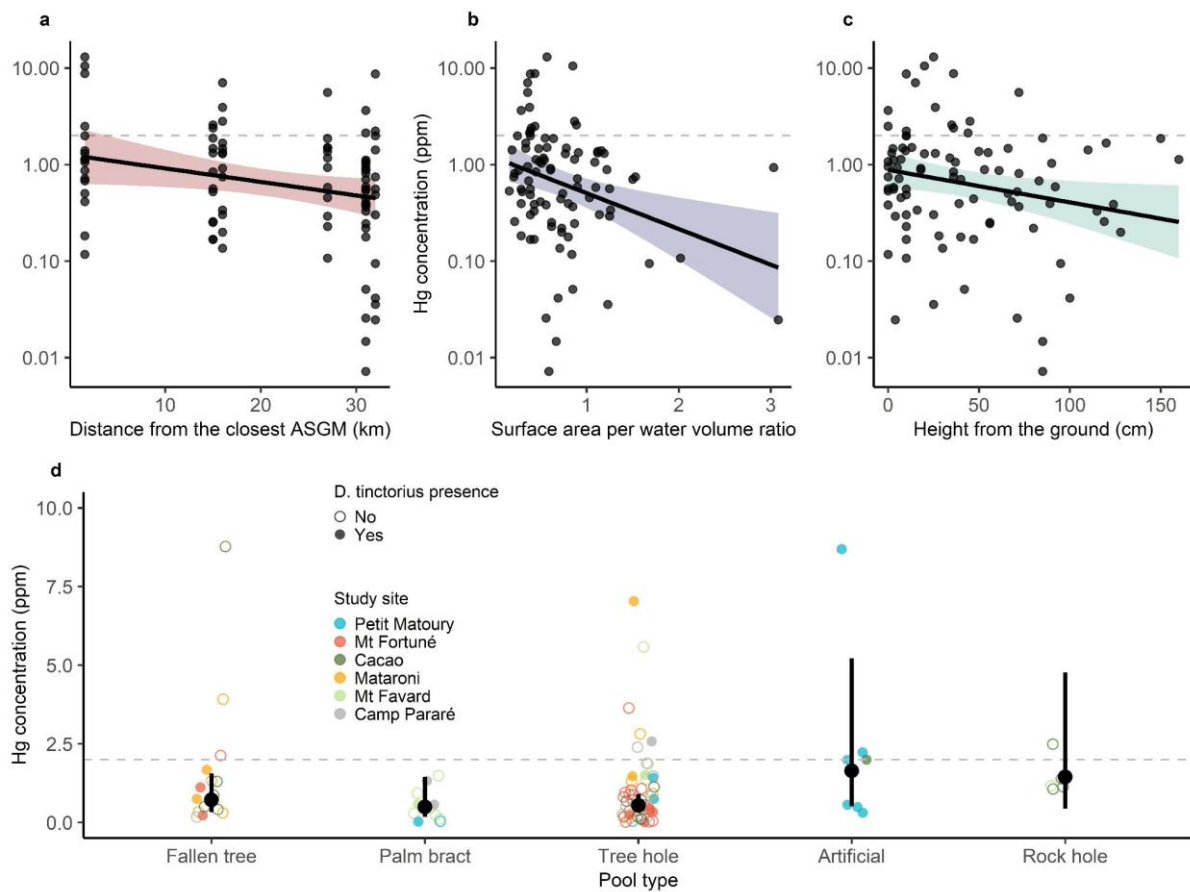


Figure 2. The predicted (large symbols and lines) Hg concentration (ppm) for (a) the distance to the closest known ASGM, (b) the surface area per water volume ratio (SA/V), (c) height above ground, and (d) the different types of phytotelmata (fallen trees, palm bracts, and tree holes) and other pool types (artificial pools and rock holes). In plot d, each colour represents one study site and filled/empty dots represent presence/absence of *D. tinctorius* tadpoles in the pool. The dashed line represents the severe effect level (2 ppm); Hg concentrations above this are considered to be heavily polluted. The 95% confidence intervals are shown as shading (a-c) or lines (d). Note that for plots a-c, the y-axis is on the log-scale for easier visualisation.

Artificial pools were common in the disturbed sites of Cacao and Petit Matoury, and often used by *Dendrobates tinctorius* as tadpole-rearing sites. No other species was recorded using this pool type. Although anthropogenic litter accumulated in disturbed areas increases the availability of pools for tadpole deposition, they could potentially act as evolutionary traps if the higher Hg levels recorded, compared to most phytotelmata, negatively affect tadpole development and/or survival. For instance, the highest Hg concentration in the presence of a *D. tinctorius* tadpole was measured in an artificial pool (8.68 ppm). Furthermore, in contrast to other studies where frogs have shown to modify their oviposition behaviour to avoid pesticides, presumably by detecting chemical pesticide cues (Takahashi 2007; Vonesh and Buck 2007), we did not observe any differences in the presence/absence or number of *D. tinctorius* tadpoles in relation to Hg concentrations (Table S2). This is in line with previous findings indicating the surprisingly wide range of pool chemical properties that *D. tinctorius* tadpoles can withstand (Fouilloux et al. 2021), which may even extend to potentially harmful substances, and suggests that phytotelm-breeding frogs might be unable to identify and/or avoid highly Hg-polluted pools for oviposition or tadpole deposition. Some Australian species, for instance, have been shown to struggle to develop in polluted wetlands, yet adults do not seem to discriminate between polluted and unpolluted pools (Sievers et al. 2019). Alternatively, it could indicate that these frogs are selecting pools on the basis of other factors such as inter-/intraspecific competition and pool stability, as is the case in several other species (Lehtinen 2004; Summers and McKeon 2004; Brown et al. 2008; Ryan and Barry 2011; Schulte et al. 2011; Fouilloux et al. 2021). However, understanding the influence of Hg contamination on pool selection requires further research on parental decision-making under controlled conditions.

Table 3. Effect size, standard error (SE), lower (LCI) and upper (UCI) 95% confidence intervals, and P-values of explanatory variables for the analysis of Hg concentrations in pools. We performed model averaging of best models ($\Delta AICc < 4$) to estimate the effect size of each variable. Informative parameters are presented in bold (95% confidence intervals do not overlap with zero).

Variable	Estimate	SE	LCI	UCI	P-value
Intercept	-0.33	0.34	-1.00	0.35	0.34
Pool height	-0.29	0.14	-0.57	-0.01	0.04
SA/V	-0.40	0.16	-0.72	-0.09	0.01
Distance from the closest ASGM	-0.37	0.21	-0.78	0.05	0.09
Pool type Palm bract	-1.07	0.71	-2.48	0.33	0.13
Pool type Tree hole	-0.27	0.40	-1.05	0.52	0.51
Pool type Artificial	0.78	0.74	-0.68	2.24	0.30
Pool type Rock hole	0.59	0.66	-0.72	1.89	0.38
pH	-0.10	0.14	-0.37	0.18	0.49

When accounting for all species recorded within the pools, which included *Allobates femoralis*, *Rhinella spp.*, and *Osteocephalus oophagus*, we found a decrease in tadpole number (but not in presence probability) with increasing Hg concentrations (Table S3; Figure 3a). However, the number of tadpoles was significantly higher in palm bracts (Table S3), consistent with the restrictions of these species to lower pools for egg/tadpole deposition compared to *D. tinctorius* (Fouilloux et al. 2021). Thus, the selection of low polluted pools may be due to a biological restriction to palm bracts, which showed lower Hg levels compared to other pool types, rather than an active avoidance of pools with higher Hg levels. In addition, the number of tadpoles also increased in pools closer to the ground, with smaller SA/V and higher pH (Table S3).

From all the tadpoles found, we sampled a total of 34 *D. tinctorius* tadpoles from developmental stages 25-29, 12 individuals from stages 30-35 and 8 from stages 36-43. This

reduction in the number of tadpoles encountered with increasing developmental stages might be due to chance (as we sampled relatively few tadpoles per pool), detection bias due to increased flight behaviour (Warkentin 1999), or mortality over time due to predation or mercury bioaccumulation. Further research should explore whether this pattern could also be explained by a lower survival of tadpoles to later stages as a consequence of longer exposure times to high mercury levels, with detrimental effects on their survival, or antipredatory behaviour. For example, Lefcort et al. (1998) reported reduced predator avoidance behaviour of tadpoles exposed to different heavy metals.

Tadpole body condition was best explained by the intercept only model (Table S4). However, there was a visual tendency for a negative correlation between Hg concentration and body condition of tadpoles at later developmental stages (Figure 3b). In addition, when excluding from our model early stage tadpoles, which have been exposed to mercury for a few weeks only (Mean \pm SD = 4.62 \pm 0.99; C. Fouilloux and B. Rojas, unpublished), mercury was included in the second-highest ranking model within Δ AIC <4 (Table S4). After model averaging, there was a negative correlation of Hg and tadpole body condition (estimate \pm SE = -0.03 \pm 0.01, Z = 3.47, p = <0.001; Table S5). More measurements from a wider range of Hg concentrations, especially of older tadpoles, would be needed to further investigate this pattern.

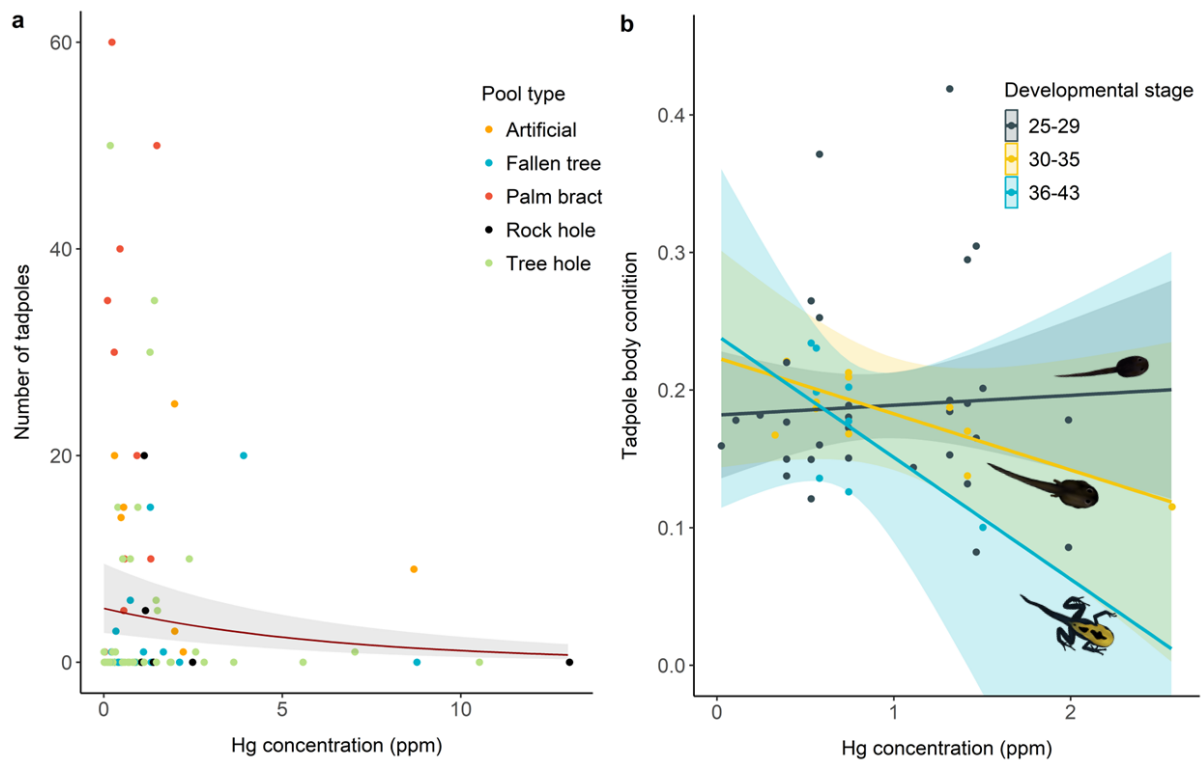


Figure 3. (a) The predicted number (line) of tadpoles from all species in relation to Hg concentration (ppm) and (b) predicted body condition of *Dendrobates tinctorius* tadpoles (lines) in relation to the Hg concentration (ppm), shown for the different developmental stage categories. Note that in this analysis, one tadpole outlier (stage 25-29) found in a pool with Hg concentration of 7.03 ppm was removed from our model. Raw data are represented as dots and 95% confidence intervals as shading.

One question that arises from this study is how much of the Hg detected in phytotelmata is ingested and accumulated in the tissues of developing tadpoles, as well as which are the potential repercussions on their development, ecology and behaviour. Bank et al. (2007) found a significant correlation between Hg levels in ponds and Hg levels in tadpole tissue, with much lower concentrations than those found in our study ($1.3e^{-6}$ – $8.4e^{-6}$ ppm). In addition, they found that Hg concentrations increased with tadpole body length, pointing towards bioaccumulation of Hg as a function of age (Bank et al. 2007). These results are in line with our findings, which point towards a detrimental effect of Hg on body condition in late developmental stages. Further, Unrine et al. (2009) found adverse effects on growth and development of Southern leopard frog larvae exposed to dietary Hg in concentrations similar

to those reported here. The data from these studies suggest that the Hg concentrations detected in phytotelmata could also bioaccumulate in phytotelm-breeding frogs, negatively affecting the development at particularly sensitive developmental stages. Finally, given the neurotoxic and endocrine-disrupting nature of Hg (Tan et al. 2009; Crump and Trudeau 2009), we urge to investigate the possible repercussions not only on the development of tadpoles, but also on their behaviour. To that end, experimental studies will be crucial to quantify the negative effects of various Hg concentrations on tadpole development, physiology, behaviour, and survival under standardised conditions, and to understand the mechanisms involved without the need to account for the myriad of confounding effects (such as predation, competition, pool type, water chemistry, etc.) that impede our ability to draw stronger conclusions from wild populations. Nevertheless, our results provide valuable baseline data of mercury concentrations in the wild and lay the foundation for future work on this important environmental issue.

CRedit authorship contribution statement

Lia Schlippe Justicia: Conceptualization, Methodology, Investigation, Data curation, Formal analysis, Visualization, Writing – original draft. **Jérémy Lemaire:** Conceptualization, Methodology, Formal analysis, Writing – review & editing. **Carolin Dittrich:** Investigation, Formal analysis, Writing – review & editing. **Martin Mayer:** Investigation, Formal analysis, Writing – review & editing. **Paco Bustamante:** Resources, Writing – review & editing. **Bibiana Rojas:** Conceptualization, Methodology, Supervision, Funding acquisition, Resources, Writing – review & editing.

Declaration of competing interest

The authors have no relevant financial or non-financial interests to disclose.

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Data availability

Data will be deposited in the Figshare repository upon acceptance.

References

Ali, H., Khan, E., & Ilahi, I. (2019). Environmental chemistry and ecotoxicology of hazardous heavy metals: environmental persistence, toxicity, and bioaccumulation. *Journal of Chemistry*, 2019, 6730305. <https://doi.org/10.1155/2019/6730305>

- Alvarez-Berríos, N. L., & Aide, T. M. (2015). Global demand for gold is another threat for tropical forests. *Environmental Research Letters*, 10(1), 014006. <https://doi.org/10.1088/1748-9326/10/1/014006>
- Alvarez-Berríos, N., Campos-Cerqueira, M., Hernández-Serna, A., Amanda Delgado, C. J., Román-Dañobeytia, F., & Aide, T. M. (2016). Impacts of small-scale gold mining on birds and anurans near the Tambopata Natural Reserve, Peru, assessed using passive acoustic monitoring. *Tropical Conservation Science*, 9(2), 832–851. <https://doi.org/10.1177/194008291600900216>
- Ashe, K. (2012). Elevated mercury concentrations in humans of Madre de Dios, Peru. *PLoS One* 7, 1–6. <https://doi.org/10.1371/journal.pone.0033305>
- Bank, M. S., Crocker, J., Connery, B., & Amirbahman, A. (2007). Mercury bioaccumulation in green frog (*Rana clamitans*) and bullfrog (*Rana catesbeiana*) tadpoles from Acadia National Park, Maine, USA. *Environmental Toxicology and Chemistry: An International Journal*, 26(1), 118–125. <https://doi.org/10.1897/07-035R.1>
- Bavec, Š., Biester, H., & Gosar, M. (2014). Urban sediment contamination in a former Hg mining district, Idrija, Slovenia. *Environmental geochemistry and health*, 36, 427-439. <https://doi.org/10.1007/s10653-013-9571-6>
- Bax, V., Francesconi, W., & Delgado, A. (2019). Land-use conflicts between biodiversity conservation and extractive industries in the Peruvian Andes. *Journal of Environmental Management*, 232, 1028–1036. <https://doi.org/10.1016/j.jenvman.2018.12.016>
- Boudou, A., Dominique, Y., Cordier, S., & Frery, N. (2006). Les chercheurs d'or et la pollution par le mercure en Guyane française: conséquences environnementales et sanitaires. *Environnement, Risques & Santé*, 5(3), 167–179.
- Brito, B. C., Forsberg, B. R., Kasper, D., Amaral, J. H., de Vasconcelos, M. R., de Sousa, O. P., Cunha, F.A., & Bastos, W. R. (2017). The influence of inundation and lake morphometry on the dynamics of mercury in the water and plankton in an Amazon floodplain lake. *Hydrobiologia*, 790, 35–48. <https://doi.org/10.1007/s10750-016-3017-y>

- Brown, J. L., Morales, V., & Summers, K. (2008). Divergence in parental care, habitat selection and larval life history between two species of Peruvian poison frogs: an experimental analysis. *Journal of Evolutionary Biology*, 21(6), 1534–1543. <https://doi.org/10.1111/j.1420-9101.2008.01609.x>
- Buchman, M. F. (2008). NOAA screening quick reference tables, NOAA OR&R Report 08-1. *Office of Response and Restoration Division, National Oceanic and Atmospheric Administration, Seattle*, pp. 34.
- Burger, J., & Snodgrass, J. (2001). Metal levels in Southern leopard frogs from the Savannah River Site: location and body compartment effects. *Environmental research*, 86(2), 157–166. <https://doi.org/10.1006/enrs.2001.4245>
- Byrne, A. R., Kosta, L., & Stegnar, P. (1975). The occurrence of mercury in amphibia. *Environmental Letters*, 8(2), 147–155. <https://doi.org/10.1080/00139307509437428>
- Campos, R. E., & Fernández, L. A. (2011). Coleopterans associated with plants that form phytotelmata in subtropical and temperate Argentina, South America. *Journal of Insect Science*, 11(1), 147. <https://doi.org/10.1673/031.011.14701>
- Carvajal-Castro, J. D., Vargas-Salinas, F., Casas-Cardona, S., Rojas, B., & Santos, J. C. (2021). Aposematism facilitates the diversification of parental care strategies in poison frogs. *Scientific Reports*, 11(1), 19047. <https://doi.org/10.1038/s41598-021-97206-6>
- Chen, H., Li, Y., Wu, H., & Li, F. (2013). Characteristics and risk assessment of heavy metals pollution of farmland soils relative to type of land use. *Journal of Ecology and Rural Environment*, 29(2), 164-169.
- Crump, K. L., & Trudeau, V. L. (2009). Mercury-induced reproductive impairment in fish. *Environmental Toxicology and Chemistry: An International Journal*, 28(5), 895–907. <https://doi.org/10.1897/08-151.1>
- Cuellar-Valencia, O. M., Murillo-García, O. E., RodríguezSalazar, G. A., & Bolívar-García, W. (2023). Bioaccumulation of mercury in direct-developing frogs: the aftermath of illegal gold mining in a National Park. *Herpetological Journal*, 33(1), 6–13. <https://doi.org/10.33256/33.1.613>

- Degetto, S., Schintu, M., Contu, A., & Sbrignadello, G. (1997). Santa Gilla lagoon (Italy): a mercury sediment pollution case study. Contamination assessment and restoration of the site. *Oceanographic Literature Review*, 2(45), 391. [https://doi.org/10.1016/S0048-9697\(97\)00165-4](https://doi.org/10.1016/S0048-9697(97)00165-4)
- Durán, A. P., Rauch, J., & Gaston, K. J. (2013). Global spatial coincidence between protected areas and metal mining activities. *Biological Conservation*, 160, 272–278. <https://doi.org/10.1016/j.biocon.2013.02.003>
- de Oliveira Drummond, L., Meire, R. O., Braga, C., de Rezende, C. E., Malm, O., & Cerqueira, R. (2022). Trophic position, altitudinal distribution, and water dependence as determining factors for mercury concentrations in tropical montane anurans. *Science of the Total Environment*, 806, 151356. <https://doi.org/10.1016/j.scitotenv.2021.151356>
- Fadini, P. S., & Jardim, W. F. (2001). Is the Negro River Basin (Amazon) impacted by naturally occurring mercury? *Science of the Total Environment*, 275(1-3), 71–82. [https://doi.org/10.1016/S0048-9697\(00\)00855-X](https://doi.org/10.1016/S0048-9697(00)00855-X)
- Fincke, O. M. (1999) Organization of predator assemblages in Neotropical tree holes: effects of abiotic factors and priority. *Ecological Entomology*, 24(1), 13–23. <https://doi.org/10.1046/j.1365-2311.1999.00166.x>
- Fisher, J. A., Schneider, L., Fostier, A. H., Guerrero, S., Guimarães, J. R. D., Labuschagne, C., Leaner, J. J., Martin, L. G., Mason, R. P., Somerset, V., & Walters, C. (2023). A synthesis of mercury research in the Southern Hemisphere, part 2: Anthropogenic perturbations. *Ambio*, 52(5), 918-937. <https://doi.org/10.1007/s13280-023-01840-5>
- Fouilloux, C. A., Serrano Rojas, S. J., Carvajal-Castro, J. D., Valkonen, J. K., Gaucher, P., Fischer, M. T., Pašukonis, A., & Rojas, B. (2021). Pool choice in a vertical landscape: tadpole-rearing site flexibility in phytotelm-breeding frogs. *Ecology and Evolution*, 11(13), 9021–9038. <https://doi.org/10.1002/ece3.7741>
- Gerson, J.R., Szponar, N., Zambrano, A.A. *et al.* (2022). Amazon forests capture high levels of atmospheric mercury pollution from artisanal gold mining. *Nature Communications*, 13, 559. <https://doi.org/10.1038/s41467-022-27997-3>

- Gibb, H., & O'Leary, K. G. (2014). Mercury exposure and health impacts among individuals in the artisanal and small-scale gold mining community: a comprehensive review. *Environmental health perspectives*, 122(7), 667–672. <https://doi.org/10.1289/ehp.1307864>
- Gomes, D. F., Moreira, R. A., Sanches, N. A. O., do Vale, C. A., Daam, M. A., Gorni, G. R., & Bastos, W. R. (2020). Dynamics of (total and methyl) mercury in sediment, fish, and crocodiles in an Amazonian Lake and risk assessment of fish consumption to the local population. *Environmental monitoring and assessment*, 192, 1–10. <https://doi.org/10.1007/s10661-020-8066-z>
- Gosner, K. L. (1960). A simplified table for staging anuran embryos and larvae with notes on identification. *Herpetologica*, 16(3), 183–190.
- Grant, T., Frost, D. R., Caldwell, J. P., Gagliardo, R. O. N., Haddad, C. F., Kok, P. J., Means, D.B., Noonan, B.P., Schargel, W.E., & Wheeler, W. C. (2006). Phylogenetic systematics of dart-poison frogs and their relatives (Amphibia: Athesphatanura: Dendrobatidae). *Bulletin of the American Museum of Natural History*, 2006(299), 1–262. [https://doi.org/10.1206/0003-0090\(2006\)299\[1:PSODFA\]2.0.CO;2](https://doi.org/10.1206/0003-0090(2006)299[1:PSODFA]2.0.CO;2)
- Grenyer, R., Orme, C. D. L., Jackson, S. F. *et al.* (2006). Global distribution and conservation of rare and threatened vertebrates. *Nature*, 444(7115), 93–96. <https://doi.org/10.1038/nature05237>
- Grimaldi, M., Guédron, S & Grimaldi, C. (2015). Impact of gold mining on mercury contamination and soil degradation in Amazonian ecosystems of French Guiana. In: F. Q. Brearley and A. D. Thomas (Eds.), *Land-Use Change Impacts on Soil Processes: Tropical and Savannah Ecosystems*. CABI. pp. 95–107.
- Heying, H. E. (2004). Reproductive limitation by oviposition site in a treehole breeding Madagascan poison frog (*Mantella laevigata*). In: R.M. Lethinen (ed.), *Ecology and Evolution of phytotelm breeding anurans*. Miscellaneous Publications, Museum of Zoology, University of Michigan, 193. pp. 23–30.

- Jardine, T. D., Kidd, K. A., & O'Driscoll, N. (2013). Food web analysis reveals effects of pH on mercury bioaccumulation at multiple trophic levels in streams. *Aquatic toxicology*, 132, 46–52. <https://doi.org/10.1016/j.aquatox.2013.01.013>
- Jiang, G. B., Shi, J. B., & Feng, X. B. (2006). Mercury pollution in China: an overview of the past and current sources of the toxic metal. *Environmental Science & Technology*, 40(12), 3672–3678. <https://doi.org/10.1021/es062707c>
- Kam, Y. C., Chuang, Z. S., & Yen, C. F. (1996). Reproduction, oviposition-site selection, and tadpole oophagy of an arboreal nester, *Chirixalus eiffingeri* (Rhacophoridae), from Taiwan. *Journal of Herpetology*, 30(1), 52–59. <https://doi.org/10.2307/1564706>
- Kidd, K., Clayden, M., & Jardine, T. (2012). Bioaccumulation and biomagnification of mercury through food webs. In: Guangliang Liu, Yong Cai, Nelson O'Driscoll (Eds.) *Environmental Chemistry and Toxicology of Mercury*. John Wiley & Sons, Inc. pp. 455-500. <https://doi.org/10.1002/9781118146644.ch14>
- Kitching, R. L. (2000). *Food webs and container habitats: the natural history and ecology of phytotelmata*. Cambridge University Press. pp. 448.
- Kumar, A., Divoll, T. J., Ganguli, P. M., Trama, F. A., & Lamborg, C. H. (2018). Presence of artisanal gold mining predicts mercury bioaccumulation in five genera of bats (Chiroptera). *Environmental Pollution*, 236, 862–870. <https://doi.org/10.1016/j.envpol.2018.01.109>
- Laurance, W.F., Vasconcelos, H.L., & Lovejoy, T.E. (2000), Forest loss and fragmentation in the Amazon: implications for wildlife conservation. *Oryx*, 34, 39–45. <https://doi.org/10.1046/j.1365-3008.2000.00094.x>
- Lawrence, J. P., Rojas, B., Fouquet, A., Mappes, J., Blanchette, A., Saporito, R.A., Bosque, R.J., Courtois, E.A., & Noonan, B.P. (2019). Weak warning signals can persist in the absence of gene flow. *Proceedings of the National Academy of Sciences*, 116(38), 19037–19045. <https://doi.org/10.1073/pnas.1901872116>
- Lefcort, H., Meguire, R. A., Wilson, L. H., & Ettinger, W. F. (1998). Heavy metals alter the survival, growth, metamorphosis, and antipredatory behavior of Columbia spotted frog

- (*Rana luteiventris*) tadpoles. *Archives of Environmental Contamination and Toxicology*, 35, 447–456. <https://doi.org/10.1007/s002449900401>
- Lehtinen, R. M. (2004). Tests for competition, cannibalism, and priority effects in two phytotelm-dwelling tadpoles from Madagascar. *Herpetologica*, 60(1), 1–13. <https://www.jstor.org/stable/3893566>
- Lehtinen, R. M., Lannoo, M. J., & Wassersug, R. J. (2004). Phytotelm-breeding anurans: past, present and future research. In: R.M. Lehtinen (ed.), *Ecology and Evolution of phytotelm breeding anurans*. Miscellaneous Publications, Museum of Zoology, University of Michigan, 193. pp. 1–9.
- Lima, F. R., Pereira, P., Junior, E. C. S., Vasques, I. C., Oliveira, J. R., Windmüller, C. C., Inda, A.V., Weindorf, D.C., Curi, N., Ribeiro, B.T., Guilherme, L.R., & Marques, J. J. (2022). Geochemistry signatures of mercury in soils of the Amazon rainforest biome. *Environmental Research*, 215, 114147. <https://doi.org/10.1016/j.envres.2022.114147>
- Malm, O., de Freitas Fonseca, M., Miguel, P. H., Bastos, W. R., & Pinto, F. N. (1998). Use of epiphyte plants as biomonitors to map atmospheric mercury in a gold trade center city, Amazon, Brazil. *Science of the Total Environment*, 213(1–3), 57–64. [https://doi.org/10.1016/S0048-9697\(98\)00074-6](https://doi.org/10.1016/S0048-9697(98)00074-6)
- Markham, K. E. & Sangermano, F. (2018). Evaluating wildlife vulnerability to mercury pollution from artisanal and small-scale gold mining in Madre de Dios, Peru. *Tropical Conservation Science*, 11, 194008291879432. <https://doi.org/10.1177/1940082918794320>
- Mason, R. P., Reinfelder, J. R., & Morel, F. M. (1996). Uptake, toxicity, and trophic transfer of mercury in a coastal diatom. *Environmental Science & Technology*, 30(6), 1835–1845. <https://doi.org/10.1021/es950373d>
- Mauro, J. B., Guimarães, J. R., & Melamed, R. (1999). Mercury methylation in a tropical macrophyte: influence of abiotic parameters. *Applied organometallic chemistry*, 13(9), 631–636. [https://doi.org/10.1002/\(SICI\)1099-0739\(199909\)13:9](https://doi.org/10.1002/(SICI)1099-0739(199909)13:9)

- Mélières, M. A., Pourchet, M., Charles-Dominique, P., & Gaucher, P. (2003). Mercury in canopy leaves of French Guiana in remote areas. *Science of the Total Environment*, 311(1–3), 261–267. [https://doi.org/10.1016/S0048-9697\(03\)00142-6](https://doi.org/10.1016/S0048-9697(03)00142-6)
- Mirlean, N., Baisch, P., Machado, I., & Shumilin, E. (2008). Mercury contamination of soil as the result of long-term phosphate fertilizer production. *Bulletin of environmental contamination and toxicology*, 81, 305-308. <https://doi.org/10.1007/s00128-008-9480-z>
- Mol, J. H., Ramlal, J. S., Lietar, C., & Verloo, M. (2001). Mercury contamination in freshwater, estuarine, and marine fishes in relation to small-scale gold mining in Suriname, South America. *Environmental Research*, 86(2), 183–197. <https://doi.org/10.1006/enrs.2001.4256>
- Mortvedt, J. J. (1995). Heavy metal contaminants in inorganic and organic fertilizers. *Fertilizer Research*, 43, 55–61. <https://doi.org/10.1007/BF00747683>
- Noonan, B. P., & Gaucher, P. (2006). Refugial isolation and secondary contact in the dyeing poison frog *Dendrobates tinctorius*. *Molecular ecology*, 15(14), 4425–4435. <https://doi.org/10.1111/j.1365-294X.2006.03074.x>
- Orr, A. G. (1994). Life histories and ecology of Odonata breeding in phytotelmata in Bornean rainforest. *Odonatologica*, 23(4), 365–377.
- Palacios-Torres, Y., Caballero-Gallardo, K., & Olivero-Verbel, J. (2018). Mercury pollution by gold mining in a global biodiversity hotspot, the Choco biogeographic region, Colombia. *Chemosphere*, 193, 421–430. <https://doi.org/10.1016/j.chemosphere.2017.10.160>
- Peig, J., & Green, A. J. (2009). New perspectives for estimating body condition from mass/length data: the scaled mass index as an alternative method. *Oikos*, 118(12), 1883–1891. <https://doi.org/10.1111/j.1600-0706.2009.17643.x>
- Peig, J., & Green, A. J. (2010). The paradigm of body condition: a critical reappraisal of current methods based on mass and length. *Functional Ecology*, 24(6), 1323–1332. <https://doi.org/10.1111/j.1365-2435.2010.01751.x>

- Peignier, M., Ringler, M., & Ringler, E. (2023). Odor cues rather than personality affect tadpole deposition in a neotropical poison frog. *Current zoology*, zoad042. <https://doi.org/10.1093/cz/zoad042>
- Quaranta, A., Bellantuono, V., Cassano, G., & Lippe, C. (2009). Why amphibians are more sensitive than mammals to xenobiotics. *PLoS One*, 4(11), e7699. <https://doi.org/10.1371/journal.pone.0007699>
- Rahm, M., Jullian, B., Lauger, A., de Carvalho, R., Vale, L., Totaram, J., Cort, K.A., Djodjodikromo, M., Hardjoprajitno, M., Neri, S., Vieira, R., Watanabe, E., do Carmo Brito, M., Miranda, P., Paloeng, C., Moe Soe Let, V., Crabbe, S., & Calmel, M. (2015). Monitoring the Impact of Gold Mining on the Forest Cover and Freshwater in the Guiana Shield. Reference year 2014. REDD+ for the Guiana Shield Project and WWF Guianas. Pp.60 https://lps16.esa.int/posterfiles/paper1306/Gold_mining_final_report_EN_vfinale.pdf
- Rice, K. M., Walker Jr, E. M., Wu, M., Gillette, C., & Blough, E. R. (2014). Environmental mercury and its toxic effects. *Journal of preventive Medicine and Public health*, 47(2), 74. <https://doi.org/10.3961/jpmph.2014.47.2.74>
- Rissoli, R. Z., Abdalla, F. C., Costa, M. J., Rantin, F. T., McKenzie, D. J., & Kalinin, A. L. (2016). Effects of glyphosate and the glyphosate based herbicides Roundup Original® and Roundup Transorb® on respiratory morphophysiology of bullfrog tadpoles. *Chemosphere*, 156, 37–44. <https://doi.org/10.1016/j.chemosphere.2016.04.083>
- Rojas, B. (2014). Strange parental decisions: fathers of the dyeing poison frog deposit their tadpoles in pools occupied by large cannibals. *Behavioral Ecology and Sociobiology*, 68, 551–559. <https://doi.org/10.1007/s00265-013-1670-y>
- Rojas, B. (2015). Mind the gap: treefalls as drivers of parental trade-offs. *Ecology and Evolution*, 5(18), 4028–4036. <https://doi.org/10.1002/ece3.1648>
- Rojas, B., & Endler, J. A. (2013). Sexual dimorphism and intra-populational colour pattern variation in the aposematic frog *Dendrobates tinctorius*. *Evolutionary Ecology*, 27, 739–753. <https://doi.org/10.1007/s10682-013-9640-4>

- Rojas, B., & Pašukonis, A. (2019). From habitat use to social behavior: natural history of a voiceless poison frog, *Dendrobates tinctorius*. *PeerJ*, 7, e7648. <https://doi.org/10.7717/peerj.7648>
- Roulet, M., Lucotte, M., Saint-Aubin, A., Tran, S., Rheault, I., Farella, N., Dezencourt, J., Passos, C.J.S., Soares, G.S., Guimarães, J.R., Mergler, D., & Amorim, M. (1998). The geochemistry of mercury in central Amazonian soils developed on the Alter-do-Chao formation of the lower Tapajos River Valley, Para state, Brazil. *Science of the Total Environment*, 223(1), 1–24. [https://doi.org/10.1016/S0048-9697\(98\)00265-4](https://doi.org/10.1016/S0048-9697(98)00265-4)
- Rowe, C. L., Hopkins, W. A., & Bridges, C. (2003). Physiological ecology of amphibians in relation to susceptibility to natural and anthropogenic factors. In: Linder, G., Krest, S., Sparling, D. (Eds.), *Amphibian Decline: An Integrated Analysis of Multiple Stressor Effects*. Pensacola: Society of Environmental Toxicology and Chemistry Press. pp. 9–58.
- Ryan, M. J., & Barry, D. S. (2011). Competitive interactions in phytotelmata—breeding pools of two poison-dart frogs (Anura: Dendrobatidae) in Costa Rica. *Journal of Herpetology*, 45(4), 438–443. <https://doi.org/10.1670/10-253.1>
- Schiesari, L., Gordo, M., & Hödl, W. (2003). Treeholes as calling, breeding, and developmental sites for the Amazonian canopy frog, *Phrynohyas resinifictrix* (Hylidae). *Copeia*, 2003(2), 263–272. [https://doi.org/10.1643/0045-8511\(2003\)003\[0263:TACBAD\]2.0.CO;2](https://doi.org/10.1643/0045-8511(2003)003[0263:TACBAD]2.0.CO;2)
- Schneider, L. (2021). When toxic chemicals refuse to die—an examination of the prolonged mercury pesticide use in Australia. *Elementa: Science of the Anthropocene*, 9(1), 053. <https://doi.org/10.1525/elementa.2021.053>
- Schulte, L. M., Yeager, J., Schulte, R., Veith, M., Werner, P., Beck, L. A., & Lötters, S. (2011). The smell of success: choice of larval rearing sites by means of chemical cues in a Peruvian poison frog. *Animal Behaviour*, 81(6), 1147–1154. <https://doi.org/10.1016/j.anbehav.2011.02.019>

- Schulte, L. M., & Lötters, S. (2014). A danger foreseen is a danger avoided: how chemical cues of different tadpoles influence parental decisions of a Neotropical poison frog. *Animal Cognition*, 17, 267-275. <https://doi.org/10.1007/s10071-013-0659-2>
- Serrano-Rojas, S. J., & Pašukonis, A. (2021). Tadpole-transporting frogs use stagnant water odor to find pools in the rainforest. *Journal of Experimental Biology*, 224(21), jeb243122. <https://doi.org/10.1242/jeb.243122>
- Shi, Q., Sun, N., Kou, H., Wang, H., & Zhao, H. (2018). Chronic effects of mercury on *Bufo gargarizans* larvae: thyroid disruption, liver damage, oxidative stress and lipid metabolism disorder. *Ecotoxicology and environmental safety*, 164, 500–509. <https://doi.org/10.1016/j.ecoenv.2018.08.058>
- Sievers, M., Hale, R., Swearer, S. E., & Parris, K. M. (2019). Frog occupancy of polluted wetlands in urban landscapes. *Conservation Biology*, 33(2), 389-402. <https://doi.org/10.1111/cobi.13210>
- Soe, P. S., Kyaw, W. T., Arizono, K., Ishibashi, Y., & Agusa, T. (2022). Mercury pollution from artisanal and small-scale gold mining in Myanmar and other southeast asian countries. *International Journal of Environmental Research and Public Health*, 19(10), 6290. <https://doi.org/10.3390/ijerph19106290>
- Song, Z., Li, P., Ding, L., Li, Z., Zhu, W., He, T., & Feng, X. (2018). Environmental mercury pollution by an abandoned chlor-alkali plant in Southwest China. *Journal of Geochemical Exploration*, 194, 81–87. <https://doi.org/10.1016/j.gexplo.2018.07.01>
- Summers, K., & McKeon, C. S. (2004). The evolutionary ecology of phytotelmata use in Neotropical poison frogs. *Miscellaneous Publications, Museum of Zoology, University of Michigan*, 193, 55–73.
- Symonds, M. R., & Moussalli, A. (2011). A brief guide to model selection, multimodel inference and model averaging in behavioural ecology using Akaike's information criterion. *Behavioral Ecology and Sociobiology*, 65, 13–21. <https://doi.org/10.1007/s00265-010-1037-6>

- Takahashi, M. (2007). Oviposition site selection: pesticide avoidance by gray treefrogs. *Environmental Toxicology and Chemistry*, 26(7), 1476–1480. <https://doi.org/10.1897/06-511R.1>
- Tan, S. W., Meiller, J. C., & Mahaffey, K. R. (2009). The endocrine effects of mercury in humans and wildlife. *Critical Reviews in Toxicology*, 39(3), 228–269. <https://doi.org/10.1080/10408440802233259>
- Tang, Z., Fan, F., Wang, X., Shi, X., Deng, S., & Wang, D. (2018). Mercury in rice (*Oryza sativa* L.) and rice-paddy soils under long-term fertilizer and organic amendment. *Ecotoxicology and Environmental Safety*, 150, 116–122. <https://doi.org/10.1016/j.ecoenv.2017.12.021>
- Turull, M., Komarova, T., Noller, B., Fontàs, C., & Díez, S. (2018). Evaluation of mercury in a freshwater environment impacted by an organomercury fungicide using diffusive gradient in thin films. *Science of the Total Environment*, 621, 1475–1484. <https://doi.org/10.1016/j.scitotenv.2017.10.081>
- UN Environment (2019). Global Mercury Assessment 2018. UN Environment Programme, Chemicals and Health Branch Geneva, Switzerland. <https://www.unep.org/resources/publication/global-mercury-assessment-2018>
- Unrine, J. M., Jagoe, C. H., Hopkins, W. A., & Brant, H. A. (2004). Adverse effects of ecologically relevant dietary mercury exposure in Southern leopard frog (*Rana sphenocephala*) larvae. *Environmental Toxicology and Chemistry: An International Journal*, 23(12), 2964–2970. <https://doi.org/10.1897/03-696.1>
- Unrine, J. M., Hopkins, W. A., Romanek, C. S., & Jackson, B. P. (2007). Bioaccumulation of trace elements in omnivorous amphibian larvae: implications for amphibian health and contaminant transport. *Environmental Pollution*, 149(2), 182–192. <https://doi.org/10.1016/j.envpol.2007.01.039>
- Varga, L. (1928). Ein interessanter Biotop der Biocönose von Wasserorganismen. *Biologisches Zentralblatt*, 48, 143–162.

- Vezzone, M., Cesar, R., Serrano, A., Lourenço, R., Rodrigues, A. P., Castilhos, Z., dos Anjos, R.M., & Polivanov, H. (2023). Mercury contamination in sediments and fish from an urban tropical estuary: ecological and human health risks. *Water, Air, & Soil Pollution*, 234(2), 72. <https://doi.org/10.1007/s11270-023-06089-y>
- Vieira, M., Bernardi, J. V., Dórea, J. G., Rocha, B. C., Ribeiro, R., & Zara, L. F. (2018). Distribution and availability of mercury and methylmercury in different waters from the Rio Madeira Basin, Amazon. *Environmental pollution*, 235, 771–779. <https://doi.org/10.1016/j.envpol.2018.01.020>
- Vonesh, J. R., & Buck, J. C. (2007). Pesticide alters oviposition site selection in gray treefrogs. *Oecologia*, 154, 219–226. <https://doi.org/10.1007/s00442-007-0811-2>
- Warkentin, K. M. (1999). The development of behavioral defenses: a mechanistic analysis of vulnerability in red-eyed tree frog hatchlings. *Behavioral Ecology*, 10(3), 251–262. <https://doi.org/10.1093/beheco/10.3.251>
- Webb, J., Coomes, O. T., Mainville, N., & Mergler, D. (2015). Mercury contamination in an indicator fish species from Andean Amazonian rivers affected by petroleum extraction. *Bulletin of environmental contamination and toxicology*, 95, 279–285. <https://doi.org/10.1007/s00128-015-1588-3>
- Weygoldt, P. (1987). Evolution of parental care in dart poison frogs (Amphibia: Anura: Dendrobatidae). *Journal of Zoological Systematics and Evolutionary Research*, 25(1), 51–67. <https://doi.org/10.1111/j.1439-0469.1987.tb00913.x>
- WHO (World Health Organization) (2017). Mercury and Health. Accessed on 03-10-2023. <https://www.who.int/news-room/fact-sheets/detail/mercury-and-health>
- Zhou, J., Obrist, D., Dastoor, A., Jiskra, M., & Ryjkov, A. (2021). Vegetation uptake of mercury and impacts on global cycling. *Nature Reviews Earth & Environment*, 2(4), 269–284. <https://doi.org/10.1038/s43017-021-00146-y>