Assessing contemporary methylmercury exposure to resident and migratory bird species of Belize

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Front cover photos: Secondary forest habitat present at the Toucan Ridge Ecology and Education Society (TREES, top), Ruddy Woodcreeper (*Dendrocincla homochroa*, bottom left), and Orange-breasted Falcon (*Falco deiroleucus*, bottom right). All photos in this report were taken by Christopher Sayers.

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Submitted on:

February 20, 2022

BIODIVERSITY RESEARCH INSTITUTE

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Suggested Citation: Sayers CJ II, Yurek H, Regan K, Evers DC. 2021. Assessing contemporary methylmercury exposure to resident and migratory bird species of Belize. Report BRI 2022-06, Biodiversity Research Institute, Portland, Maine, USA.

Abstract

Anthropogenic mercury (Hg) is a systemic environmental toxin throughout the Neotropics that may pose population-level impacts to fish- and invertebrate-eating bird species. During a rapid avian Hg assessment conducted in the wet season of 2021 by the Biodiversity Research Institute and the Foundation for Wildlife Conservation, we found 72% of well-sampled species ($n \geq 10$) had lower mean Hg concentrations than in previous surveys conducted from 2007–2019, while 80% of sites had higher mean Hg concentrations. When excluding poorly-sampled species ($n < 10$) from our most recent survey, only the American Pygmy Kingfisher (*Chloroceryle aenea*) had blood Hg concentrations that exceeded adverse effect thresholds associated with reproductive impairment. Previous Neotropical bird Hg biomonitoring efforts (Sayers et al. *In review*) highlight central Belize as a biological Hg hotspot, but this contemporary assessment may suggest that Hg biomagnification is deescalating or atmospheric Hg deposition is decreasing throughout the Yucatán Peninsula. To expand our understanding of these evolving dynamics in Belize, as a function of BRI's on-the-ground involvement, several Belizean conservation organizations are actively contributing to bird tissue collection efforts in collaboration with the Tropical Research for Avian Conservation and Ecotoxicology (TRACE) Initiative.

Background

Mercury (Hg) is a widely-distributed and persistent environmental pollutant with detrimental implications for global health. International government organizations to curtail global Hg emissions, such as the United Nations Minamata Convention on Mercury, have made transformative progress in generating the research and legislation necessary to regulate polluting industries throughout the Global North. However, such action is severely lacking in tropical nations despite rising emissions throughout the Global South. As a function of rising gold demand and price since the beginning of the century, the rapid expansion of artisanal and small-scale gold mining (ASGM) has catapulted the global tropics as the leading contributor of environmental Hg pollution (UNEP 2019). This paradigm shift is cause for major concern in how we consider and assess Hg health risks to tropical ecosystems and their inhabitants.

Tropical wet forest biomes maintain biogeochemical conditions, such as elevated precipitation and seasonal flooding, that enhance ASGM emissions by promoting Hg bioavailability. Recent work by Sayers et al. (*In review*) and Gerson et al. (2022) have begun to pinpoint which biotic and abiotic factors predispose biodiversity to high Hg exposure — using birds as accessible and economical bioindicators. Tropical Hg biomagnification is expected to intensify for 1) sites within close proximity ($\lesssim$ 2 km) to artisanal gold mining or processing zones, 2) sites with intact, robust forest canopies, 3) sites where Hg methylating habitats, such as wetlands, are abundant throughout the landscape, and 4) organisms feeding at high trophic positions. To summarize, fish- and invertebrate-eating bird species foraging within primary rainforest or wetland habitats adjacent to ASG mining or processing sites are predicted to have exceedingly high blood and feather Hg concentrations relative to the remaining avian community. There is now mounting evidence associating high Hg exposure with reproductive, behavioral, immunological, neurological, and physiological impairment in wildlife (Depew et al. 2012; Dietz et al. 2013; Scheuhammer et al. 2015), particularly birds (Evers et al. 2005; Ackerman et al. 2016; Fuchsman et al. 2017; Evers 2018; Whitney and Cristol 2018). And while these negative effects are seldom applied at the population level, Hg exposure could be a "silent" contributor to avian biodiversity loss throughout the Americas (Rosenberg et al. 2019; Stouffer et al. 2020; Sherry 2021) — highlighting the need for consistent, widespread Hg monitoring.

Since 2007, the Biodiversity Research Institute (BRI) has collaborated with numerous conservation organizations in Belize to begin understanding Hg dynamics within terrestrial tropical ecosystems — using resident and migratory bird species as key bioindicators. Preliminary objectives included developing Hg exposure profiles for terrestrial bird species, documenting geographic Hg hotspots and important methylating habitats, and communicating findings to government agencies and local communities to raise awareness about the impacts of Hg on human, wildlife, and ecosystem health. After over a decade of infrequent avian Hg sampling expeditions and the analysis of over 450 blood and feather samples, central Belize has emerged as a biological Hg hotspot, which has produced elevated concentrations across a variety of avifauna (Evers and Burton 2020; Sayers et al. *In review*).

While previous work has defined a valuable baseline from which to compare future data, the data showcased in this report represent the most recent Hg assessment for central Belize and can help illustrate how avian Hg concentrations and risk have changed over the past 15 years.

Objectives

1. Generate contemporary avian Hg exposure profiles and risk assessments
2. Quantify how avian Hg exposure and risk are changing through time
3. Identify several relatively common, broadly distributed resident bird species to serve as future sentinel Hg bioindicators across Latin America
4. Create and strengthen scientific partnerships with bird conservation organizations by holding in-person meetings and lectures

Methods

Study areas

To collect avian tissue samples for later Hg analysis, we conducted ground-level mist net surveys from August 25 through October 15, 2021 at the Tropical Education Center, Runaway Creek Nature Reserve, Monkey Bay Wildlife Sanctuary, and Toucan Ridge Ecology and Education Society with primary assistance from staff at the Foundation for Wildlife Conservation, Tropical Education Center, 28 George Price Highway, P.O. Box 368, La Democracia, Belize District, Belize. We selected study locations in a variety of habitats including riparian scrub, pine savanna, and secondary forest. Site-specific capture and habitat information are featured in Appendix 1.

Figure 1. Karst hill broadleaf forest, pine savanna, and seasonal wetland matrix at Runaway Creek Nature Reserve (left) and mist net array along Sibun River (right).

Sample Collection

We captured birds using 12 m mist nets and identified individuals to an appropriate species, sex, age, and molt cycle using a local bird field guide (Fagan and Komar 2016) and either a calendar-based, or the preferred, Wolfe-Ryder-Pyle (WRP) cycle-based age-classification system (Wolfe et al. 2010; Johnson et al. 2011; Tórrez and Arendt 2012, 2017; Pyle et al. 2016) described for Neotropical bird families in Johnson and Wolfe (2017). Whenever possible, we banded all taxa, excluding hummingbirds (Trochilidae), with an aluminum leg band from the US Fish and Wildlife Service and the National Band & Tag Company (https://www.nationalband.com/, Newport, Kentucky, USA). In unique circumstances when we did not possess aluminum leg bands in the proper size, we banded individuals with a unique combination of colored plastic legs bands and assigned the corresponding samples a unique identification number. We also measured and assessed birds for feather molt and wear, skull ossification, fat stores, muscle mass, wing chord, tarsus length, bill dimensions, tail length, body mass, and reproductive stage via cloacal protuberance and brood patch.

We followed tissue collection, preparation, and storage methods provided by the Biodiversity Research Institute (Evers et al. 2021). Whenever possible, we collected 30–60 µL of blood from the cutaneous ulnar vein using 75 µL heparinized capillary tubes, sealed tubes at both ends using Critocaps™ or Critoseal™, placed tubes into a plastic vacutainer, and stored samples in a cooler with ice packs. We transferred blood samples into a freezer within 8 hours of collection where they were stored below –4 ºC until laboratory analysis. For feather sampling, we collected the two outermost tail feathers and 5–6 flank feathers and stored samples in paper coin envelopes at ambient temperature.

While we collected multiple tissue types from each individual to obtain a holistic sample, we only showcase the whole blood data in this report. Whole blood best represents recent dietary Hg exposure from days to weeks, which makes it an ideal sampling matrix for rapid site assessments (Evers et al. 2005; Evers 2018).

Figure 2. Preparing a Green Kingfisher (*Chloroceryle americana*) for blood sampling via brachial venipuncture.

Laboratory analysis

We performed total mercury analyses at the Biodiversity Research Institute Toxicology Lab (Portland, Maine, USA) using a thermal decomposition and atomic absorption spectrophotometry technique with a Nippon MA-3000 direct Hg analyzer. We assumed that nearly all total mercury (THg, > 95%) in whole blood (Rimmer et al. 2005; Edmonds et al. 2010) to be in the bioavailable, methylmercury (MeHg) form. Therefore, all sampled tissue concentrations represent MeHg contamination of Neotropical avifauna. We followed United States Environmental Protection Agency (EPA) SW-846 Method 7473, "Mercury in solids and solutions by thermal decomposition, amalgamation, and atomic absorption spectrophotometry" (USEPA 1998), and used quality control methods (including certified reference materials DOLT-5 and ERM-CE464) to ensure consistent analytical precision and accuracy. We excluded any sample that registered below the analyzer's lower detection limit (0.05 ng THg) from the final database.

Results and Discussion

Hg exposure and risk

We present a collective database containing 546 whole blood THg samples from 7 orders, 27 families, and 86 bird species throughout central Belize collected from 2007–2021. The most well-sampled bird families included New World warblers (Parulidae; n = 170), tyrant flycatchers (Tyrannidae; n = 64), woodcreepers (Furnariidae; n = 45), tanagers (Thraupidae; n = 44), wrens (Troglodytidae; n = 35), manakins (Pipridae; n = 32), vireos (Vireonidae; n = 28), and kingfishers (Alcedinidae; n = 25).

Recent research on Neotropical bird Hg exposure documents consistently elevated Hg concentrations in bird communities of the Yucatán Peninsula from 2007–2019 and has highlighted central Belize as a biological Hg hotspot relative to other regions throughout the Neotropical realm (Sayers et al. *In review*). Belize may face elevated Hg deposition because of gaseous elemental Hg emissions from local landfill incineration, coal combustion in central Mexico (UNEP 2019), as well as industrial and artisanal gold mining in the Chiquibul/Maya Mountains (Cornec 2010; Briggs et al. 2013; Manzanero 2014; Rath 2016). In addition, there is an abundance of key Hg methylating habitats at some of the Belize sampling sites, such as seasonal wetlands at Runaway Creek Nature Reserve, that we expect can readily convert inorganic emissions to a bioavailable form.

Our contemporary Hg assessment conducted in the wet season of 2021 opposes this present school of thought by providing generally positive, but paradoxical, results for Hg biogeochemical cycling in Belize. We found that 72% of well-sampled bird species ($n \geq 10$) had lower mean Hg concentrations relative to previous surveys conducted from 2007–2019 (Fig. 1), while 80% of sites had higher mean Hg concentrations (Fig. 2). Perhaps the most valuable way of assessing these results is to put them into context of established adverse effect thresholds. In our contemporary survey, the American Pygmy Kingfisher (*Chloroceryle aenea*) was the only species that exhibited blood Hg concentrations associated with a 10% loss in reproductive success (Fig. 3; Jackson et al. 2011). This is a marked improvement compared to historical surveys, in which a total of 4 species exceeded reproductive impairment thresholds (Fig. 3). Therefore, these factors may indeed suggest that avian Hg risk and biomagnification are deescalating or atmospheric Hg deposition is decreasing throughout the Yucatán Peninsula — uplifting, but unexpected findings that are deserving of further scrutiny.

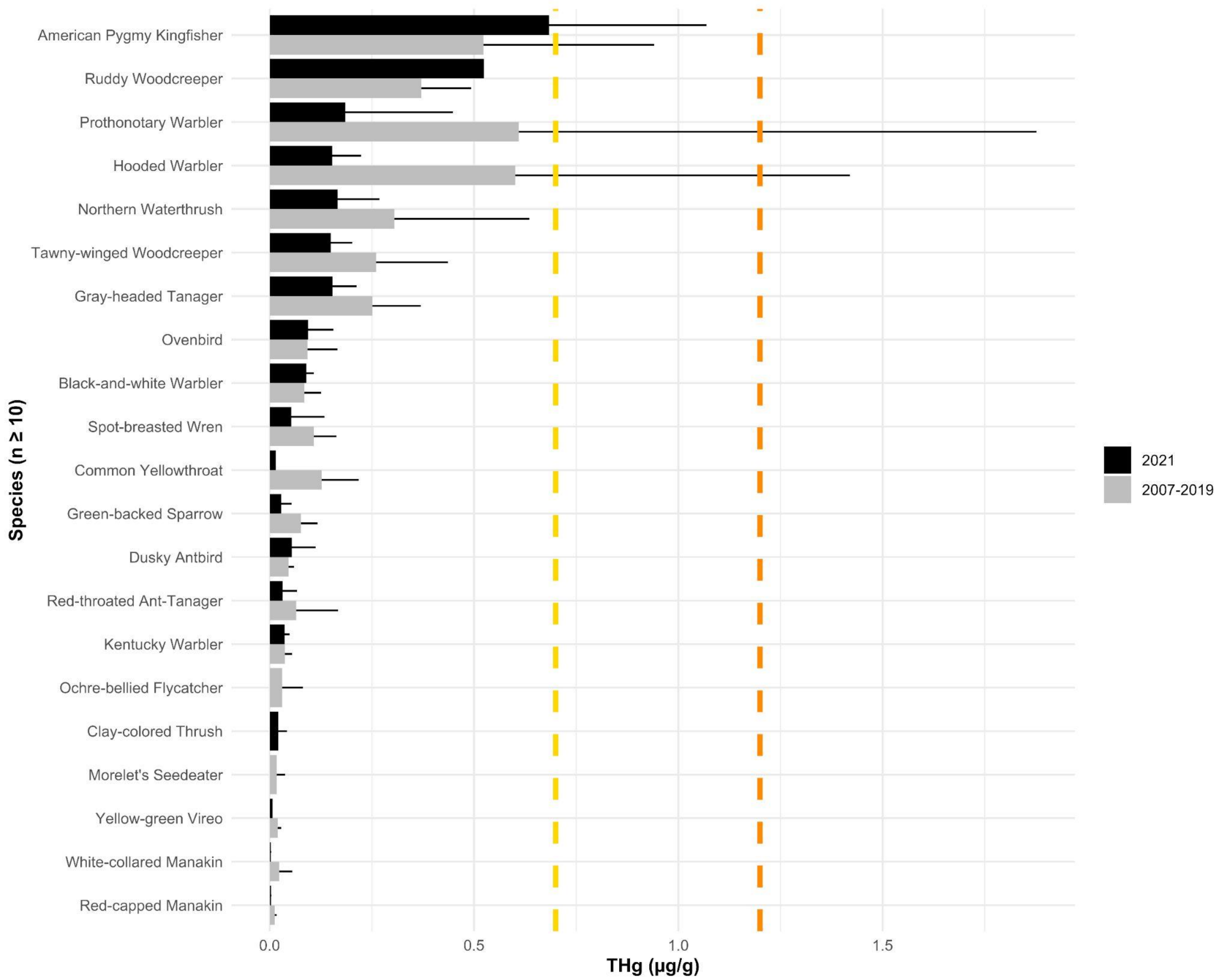

Figure 1. Arithmetic mean ± standard deviation of whole blood total mercury (THg) concentrations (µg/g) among bird species sampled throughout central Belize from 2007–2021. The dashed horizontal lines indicate risk thresholds defined by Jackson et al. (2011) where ≥ 0.7 µg/g ww represents a ≥ 10% decline in reproductive success (yellow) and ≥ 1.2 µg/g ww represents a ≥ 20% decline in reproductive success (orange).

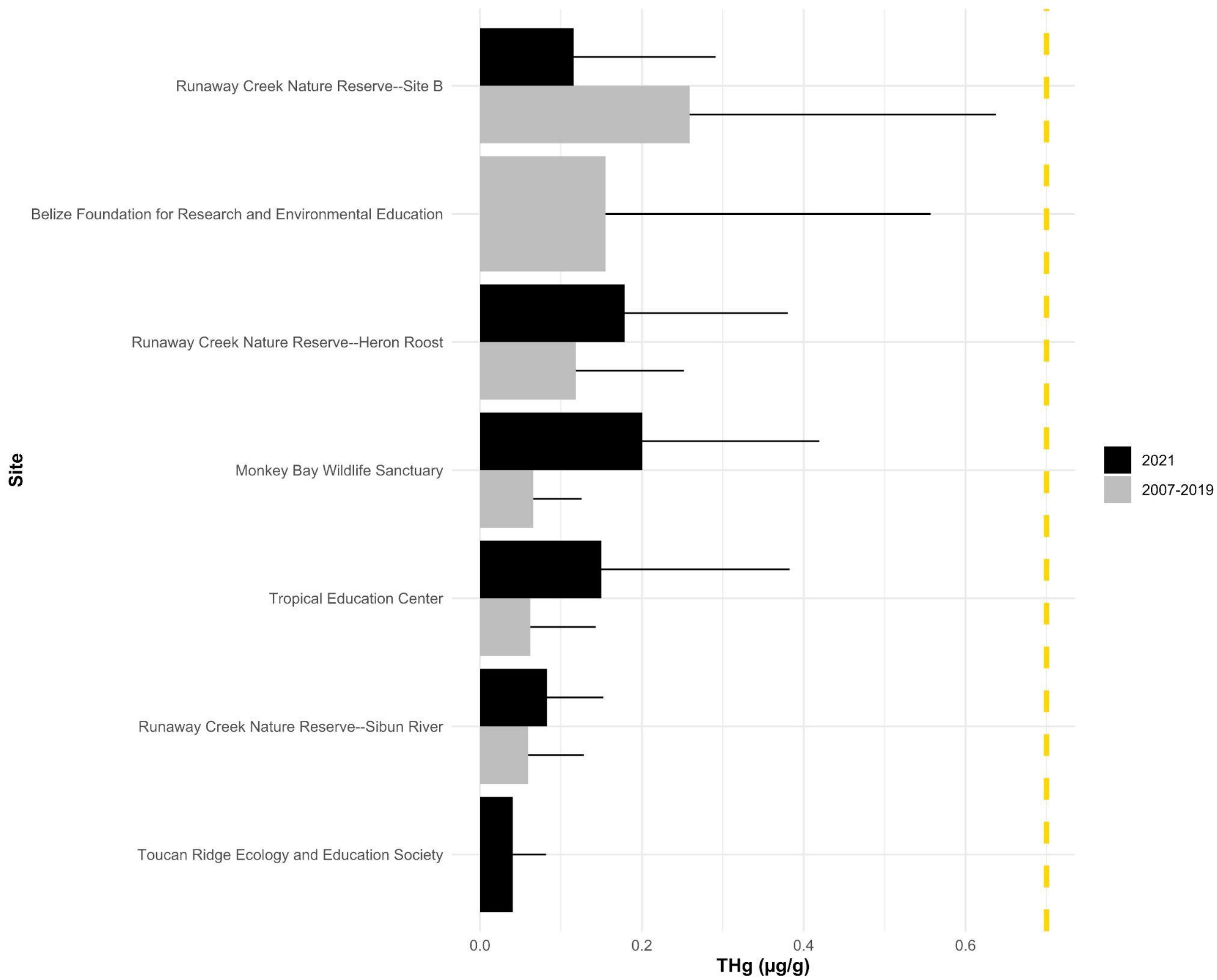

Figure 2. Arithmetic mean ± standard deviation of whole blood total mercury (THg) concentrations (µg/g) among sites sampled throughout central Belize from 2007–2021. The yellow dashed horizontal line indicates a risk threshold defined by Jackson et al. (2011) where ≥ 0.7 µg/g ww represents a ≥ 10% decline in reproductive success.

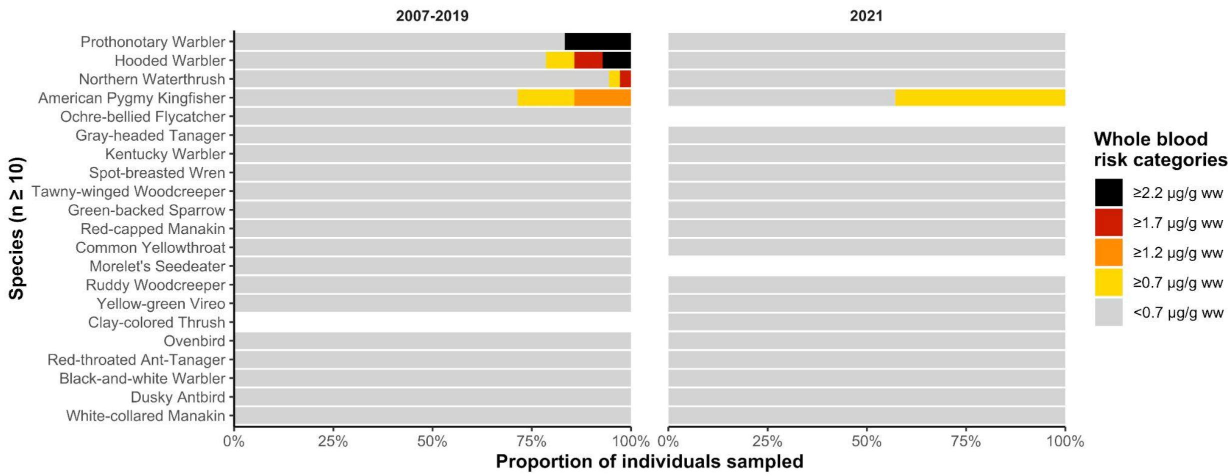

Figure 3. Proportion of Neotropical bird species sampled for whole blood throughout central Belize from 2007–2021 that may be subject to different degrees of reproductive failure via MeHg exposure. Species with fewer than 10 samples were excluded. Whole blood risk categories defined by Jackson et al. (2011) include: < 0.7 µg/g ww (gray, represents a ≤ 10% decline in reproductive success), ≥ 0.7 µg/g ww (yellow, represents a ≥ 10% decline in reproductive success), ≥ 1.2 µg/g ww (orange, represents a ≥ 20% decline in reproductive success), ≥ 1.7 µg/g ww (red, represents a ≥ 30% decline in reproductive success), and ≥ 2.2 µg/g ww (black, represents a ≥ 40% decline in reproductive success).

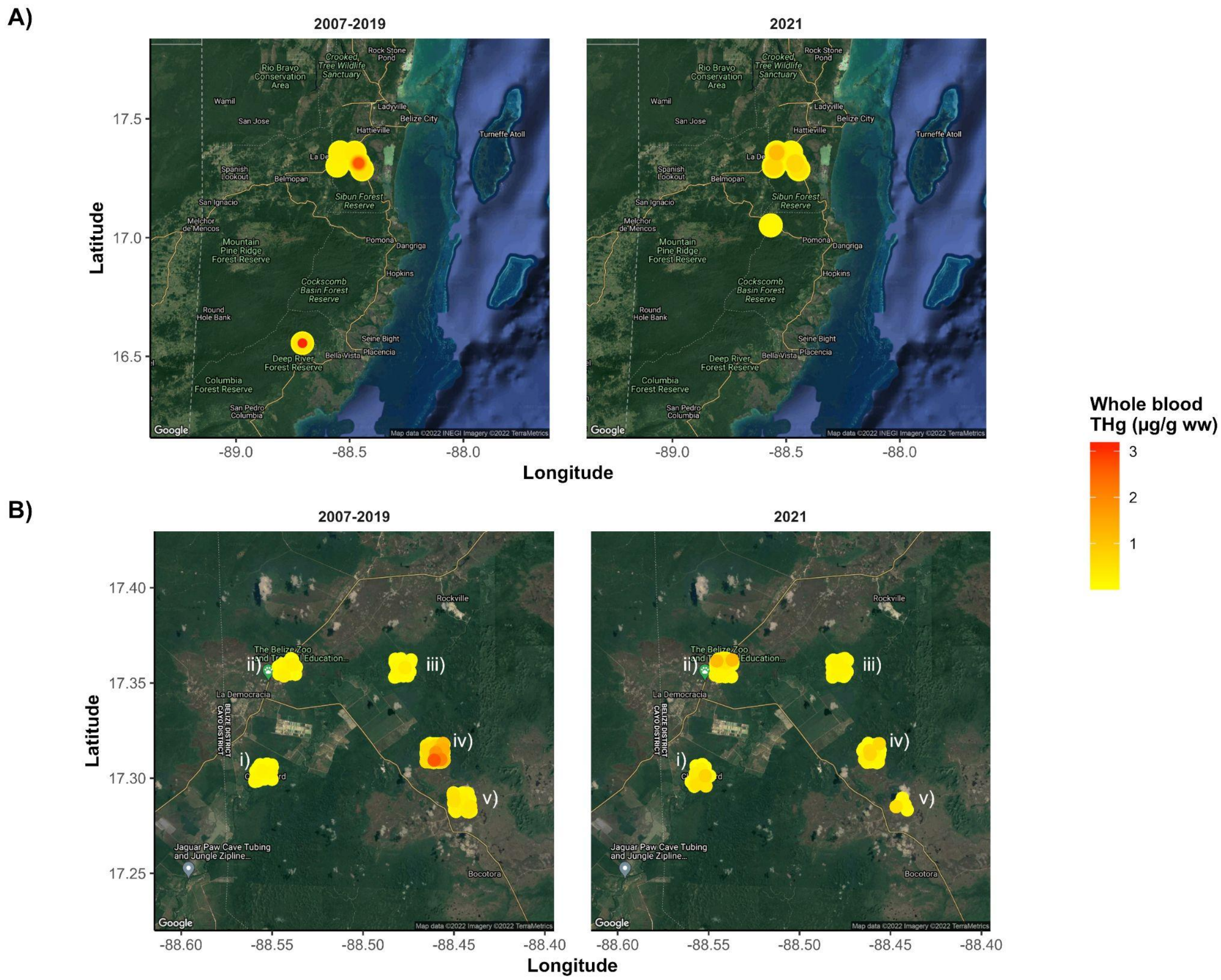

Figure 4. Map displaying total mercury (THg) concentrations (μg/g) of 546 whole blood samples from 86 bird species collected throughout central Belize collected from 2007–2021. Map **A)** displays THg concentrations on a country aggregate, while **B)** displays THg concentrations from sites i) Monkey Bay Wildlife Sanctuary, ii) Tropical Education Center, iii) Sibun River, iv) Site B, and v) Heron Roost.

Strengthening scientific collaborations

From September 27–October 1 and October 11–15, 2021, field biologists from University of Belize Environmental Research Institute, BRI, Runaway Creek Nature Reserve, Monkey Bay Wildlife Sanctuary, Belize Audubon Society, and Toledo Institute for Development and Environment (TIDE) convened at TREES for two overnight workshops to learn hands-on bird banding skills while operating an established Monitoreo de Sobrevivencia Invernal (MoSI, see https://www.birdpop.org/pages/mosi.php) station (Fig. 5). This event series provided the perfect venue to unite the majority of the Belizean bird banding community, communicate essential bird banding theory and field skills, and convey the mission and principal findings of BRI's ongoing Neotropical Hg research. Specifically, as part of his role as workshop co-instructor, Christopher Sayers gave multiple presentations on BRI's previous bird sampling throughout Central America and the Caribbean — highlighting a newly-developed collaborative opportunity for attending organizations.

To overcome previous methodological inconsistencies and inferential uncertainties in the field of tropical ecotoxicology, BRI has developed an international, collaborative data-sharing platform known as the Tropical Research for Avian Conservation and Ecotoxicology (TRACE) Initiative (see https://www.briwildlife.org/TRACE). Through the equitable transfer of funding, equipment, data, and authorship among collaborators, TRACE seeks to better inform biodiversity conservation efforts through the understanding of the prevalence, spatiotemporal distribution, toxicokinetics, and biogeochemistry of Hg throughout the tropics — using birds as a convenient, ecologically important, and charismatic proxy. Such standardized information can then be used for long-term Hg monitoring purposes to inform local, national, and international government entities, including Parties of the United Nations Minamata Convention on Mercury.

During the cumulative 10 workshop days, leaders and participants assisted Christopher in collecting over 150 blood and feather samples from resident and migratory bird species. As a result, attendees gained the tissue extraction skills necessary to contribute samples to the TRACE Initiative, as well as pursue research of their own design. As a direct result of this workshop series, BRI is actively collaborating with Foundation for Wildlife Conservation, Monkey Bay Wildlife Sanctuary, University of Belize Environmental Research Institute, Belize Audubon Society, TIDE, Corozal Sustainable Future Initiative (CSFI), and TREES to collect avian tissue samples for later Hg analysis via the TRACE Initiative. Bird Hg samples collected at their respective reserve systems will greatly expand our knowledge about the spatial distribution of Hg throughout Belize, and aid us in identifying biological hotspots across different habitats.

Figure 5. Group pictures of bird Hg sampling training held at Toucan Ridge Ecology and Education Society.

Conclusions

Given Belize's interest in ratifying the United Nations Minamata Convention on Mercury and membership of the newly organized Caribbean Region Mercury Monitoring Network (see https://briwildlife.org/hgcenter/crmmn/; Evers and Burton 2021), Belize is uniquely positioned throughout the Neotropics to serve as a long-term study area for assessing the spatiotemporal trends and impacts of Hg exposure to Neotropical birds. Study sites including the Runaway Creek Nature Reserve and TREES provide an ideal framework to build local capacity on bird banding, population science, and ecotoxicological sampling via their educational outreach programs, MoSI stations, and newly-adopted TRACE Initiative.

Recommendations

1. Continue monitoring avian Hg exposure at Runaway Creek Nature Reserve, Tropical Education Center, Monkey Bay Wildlife Sanctuary, and Toucan Ridge Ecology and Education Society as long-term study sites.
2. Expand avian Hg biomonitoring efforts to incorporate sites within the Chiquibul/Maya Mountains to assess potential biotic impacts of artisanal gold mining Hg emissions.
3. Prioritize sampling abundant and widespread invertivore and piscivore sentinel species that exhibit elevated Hg concentrations, including American Pygmy Kingfisher (*Chloroceryle aenea*), Hooded Warbler (*Setophaga citrina*), and Tawny-winged Woodcreeper (*Dendrocincla anabatina*), to broaden the taxonomic and geographic applicability of Hg data from Belize (Appendix 3).
4. Approach Hg sampling using a "food web strategy" by taking soil, leaf, ant, and arachnid samples to better understand avian Hg biomagnification dynamics using stable Hg isotope signatures.

Acknowledgements

This effort would not have been possible without staff assistance from the Foundation for Wildlife Conservation, Tropical Education Center, Monkey Bay Wildlife Sanctuary, Toucan Ridge Ecology and Education Society, and University of Belize Environmental Research Institute. We thank Reynold Cal, Stevan Reneau, Gilroy "Niko" Welch, Wilber Martínez, Kayla Hartwell, Abidas Ash, Finola Fogarty, and Mat Charette for field assistance; Kayla Hartwell, Juan Carlos, Abidas Ash, and Mat Charette for organizing logistics; and Matthew Miller and Mat Charette for granting site access. We especially thank the Biodiversity Research Institute and the Cornell Lab of Ornithology for funding this project.

Figure 6. Barred Antshrike (*Thamnophilus doliatus*) at Runaway Creek Nature Reserve.

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Appendix 1. Total mercury (THg) concentrations (µg/g) among all 7 sites sampled throughout Belize from 2007–2021. Number of species sampled, sample size (*n*), arithmetic mean ± standard deviation (SD), range, and coefficient of variation (CV) are summarized by site, habitat, and year. A dash (–) indicates there are no data to report.

Site (latitude, longitude)	Habitat	Year	Species sampled	*n*	Arithmetic mean ± SD	Range	CV
Belize Foundation for Research & Environmental Education (16.55578, –88.70777)	Shade coffee/cacao plantation; secondary forest	2007–2019	13	65	0.155 ± 0.402	0.010–3.195	258.9%
Monkey Bay Wildlife Sanctuary (17.30258, –88.55478)	River-edge forest; riparian thickets	2007–2019	11	28	0.066 ± 0.060	0.008–0.222	90.4%
		2021	12	19	0.201 ± 0.219	0.001–0.741	108.9%
Runaway Creek Nature Reserve--Heron Roost (17.287262, –88.44553)	Riparian thickets; secondary forest	2007–2019	11	54	0.118 ± 0.134	0.005–0.57	112.9%
		2021	5	5	0.179 ± 0.202	0.004–0.525	113.1%
Runaway Creek Nature Reserve--Sibun River (17.358047, –88.47801)	Tropical lowland evergreen forest; second-growth shrub	2007–2019	18	32	0.060 ± 0.068	0.005–0.37	114.3%
		2021	17	39	0.083 ± 0.070	0.003–0.234	83.9%
Runaway Creek Nature Reserve--Site B (17.313222, –88.460329)	Secondary forest; low, seasonally wet grassland	2007–2019	49	150	0.259 ± 0.379	0.003–2.659	146.4%
		2021	25	41	0.116 ± 0.176	0.002–0.719	151.9%
Toucan Ridge Ecology and Education Society (17.05231, -88.56773)	Secondary forest; tropical lowland evergreen forest	2021	14	21	0.040 ± 0.041	0.002–0.138	101.8%
Tropical Education Center (17.35812, –88.54162)	Secondary forest; pine-oak forest; gallery forest	2007–2019	19	35	0.062 ± 0.081	0.001–0.293	129.5%
		2021	23	57	0.150 ± 0.232	0.001–1.187	154.9%

Appendix 2. Whole blood total mercury (THg) concentrations (µg/g) among all 86 Neotropical resident and migratory bird species sampled throughout Belize from 2007–2021. Sample size (*n*), arithmetic mean ± standard deviation (SD), range, and coefficient of variation (CV) are summarized by family, species, and year. Species are arranged alphabetically by order and family. A dash (–) indicates there are no data to report.

Family	Common name	Latin name	Year	*n*	Arithmetic mean ± SD	Range	CV
Alcedinidae (Kingfishers)	American Pygmy Kingfisher	*Chloroceryle aenea*	2007–2019	14	0.523 ± 0.418	79.9%	0.069–1.37
			2021	7	0.683 ± 0.386	56.4%	0.159–1.187
Alcedinidae (Kingfishers)	Green Kingfisher	*Chloroceryle americana*	2007–2019	1	0.515	—	—
			2021	3	0.446 ± 0.048	10.7%	0.395–0.49
Caprimulgidae (Nightjars and Allies)	Common Pauraque	*Nyctidromus albicollis*	2021	1	0.036	—	—
Cardinalidae (Cardinals and Allies)	Blue Bunting	*Cyanocompsa parellina*	2007–2019	1	0.045	—	—
			2021	1	0.007	—	—
	Gray-throated Chat	*Granatellus sallaei*	2021	1	0.14	—	—
	Red-throated Ant-Tanager	*Habia fuscicauda*	2007–2019	9	0.065 ± 0.102	157.3%	0.017–0.337
			2021	2	0.031 ± 0.035	111.9%	0.007–0.056
Columbidae (Pigeons and Doves)	Ruddy Ground Dove	*Columbina talpacoti*	2021	1	0.001	—	—
	White-tipped Dove	*Leptotila verreauxi*	2007–2019	1	0.007	—	—
Corvidae (Crows, Jays, and Magpies)	Brown Jay	*Psilorhinus morio*	2007–2019	1	0.027	—	—
Fringillidae (Finches, Euphonias, and Allies)	Yellow-throated Euphonia	*Euphonia hirundinacea*	2007–2019	2	0.006 ± 0.002	37.0%	0.005–0.008
Furnariidae (Ovenbirds and Woodcreepers)	Ivory-billed Woodcreeper	*Xiphorhynchus flavigaster*	2021	1	0.156	—	—
	Olivaceous Woodcreeper	*Sittasomus griseicapillus*	2007–2019	2	0.112 ± 0.045	40.5%	0.08–0.144

	Plain Xenops	*Xenops minutus*	2007–2019	1	0.045	—	—
	Ruddy Woodcreeper	*Dendrocincla homochroa*	2007–2019	13	0.371 ± 0.122	32.9%	0.222–0.598
			2021	1	0.525	—	—
	Rufous-breasted Spinetail	*Synallaxis erythrothorax*	2007–2019	1	0.045	—	—
			2021	3	0.066 ± 0.059	89.9%	0.01–0.128
	Streak-headed Woodcreeper	*Lepidocolaptes souleyetii*	2007–2019	2	0.145 ± 0.017	11.8%	0.133–0.157
	Tawny-winged Woodcreeper	*Dendrocincla anabatina*	2007–2019	11	0.261 ± 0.176	67.4%	0.073–0.607
			2021	9	0.15 ± 0.053	35.3%	0.06–0.234
	Wedge-billed Woodcreeper	*Glyphorynchus spirurus*	2007–2019	1	0.278	—	—
Galbulidae (Jacamars)	Rufous-tailed Jacamar	*Galbula ruficauda*	2021	1	0.063	—	—
Hirundinidae (Swallows)	Northern Rough-winged Swallow	*Stelgidopteryx serripennis*	2007–2019	2	0.173 ± 0	0.1%	0.173–0.173
Icteridae (Troupials and Allies)	Baltimore Oriole	*Icterus galbula*	2007–2019	1	0.008	—	—
	Black-cowled Oriole	*Icterus prosthemelas*	2021	1	0.007	—	—
	Yellow-billed Cacique	*Amblycercus holosericeus*	2007–2019	3	0.112 ± 0.127	113.9%	0.032–0.259
			2021	4	0.028 ± 0.013	46.1%	0.016–0.045
Icteriidae (Yellow-breasted Chat)	Yellow-breasted Chat	*Icteria virens*	2007–2019	3	0.046 ± 0.065	142.3%	0.006–0.12
			2021	1	0.059	—	—
Mimidae (Mockingbirds and Thrashers)	Gray Catbird	*Dumetella carolinensis*	2007–2019	9	0.022 ± 0.012	52.1%	0.012–0.048
Momotidae (Motmots)	Lesson's Motmot	*Momotus lessonii*	2021	1	0.036	—	—
Oxyruncidae (Sharpbill, Royal Flycatcher, and Allies)	Royal Flycatcher	*Onychorhynchus coronatus*	2007–2019	2	0.545 ± 0.164	30.2%	0.429–0.662

			2021	1	0.014	—	—
Parulidae (New World Warblers)	American Redstart	*Setophaga ruticilla*	2007–2019	4	0.263 ± 0.133	50.4%	0.101–0.414
	Black-and-white Warbler	*Mniotilta varia*	2007–2019	4	0.085 ± 0.041	48.3%	0.033–0.13
			2021	6	0.09 ± 0.018	20.2%	0.07–0.115
	Common Yellowthroat	*Geothlypis trichas*	2007–2019	14	0.127 ± 0.091	71.3%	0.023–0.4
			2021	1	0.015	—	—
	Hooded Warbler	*Setophaga citrina*	2007–2019	14	0.601 ± 0.819	136.2%	0.047–2.659
			2021	12	0.153 ± 0.071	46.2%	0.067–0.288
	Kentucky Warbler	*Geothlypis formosa*	2007–2019	19	0.037 ± 0.017	46.1%	0.014–0.085
			2021	4	0.037 ± 0.012	31.9%	0.023–0.049
	Louisiana Waterthrush	*Parkesia motacilla*	2007–2019	2	0.399 ± 0.001	0.4%	0.398–0.4
			2021	6	0.297 ± 0.114	38.5%	0.131–0.468
	Magnolia Warbler	*Setophaga magnolia*	2007–2019	7	0.096 ± 0.055	57.6%	0.041–0.208
	Mourning Warbler	*Geothlypis philadelphia*	2021	1	0.028	—	—
	Northern Waterthrush	*Parkesia noveboracensis*	2007–2019	36	0.305 ± 0.33	108.2%	0.047–1.804
			2021	6	0.166 ± 0.102	61.5%	0.041–0.339
	Ovenbird	*Seiurus aurocapilla*	2007–2019	7	0.093 ± 0.073	79.1%	0.021–0.202
			2021	5	0.094 ± 0.062	65.6%	0.045–0.201
	Prothonotary Warbler	*Protonotaria citrea*	2007–2019	6	0.609 ± 1.267	207.9%	0.043–3.195
			2021	5	0.185 ± 0.263	142.1%	0.04–0.654
	Swainson's Warbler	*Limnothlypis swainsonii*	2021	1	0.208	—	—
	Tennessee Warbler	*Leiothlypis peregrina*	2007–2019	1	0.022	—	—
	Worm-eating Warbler	*Helmitheros vermivorum*	2007–2019	2	0.022 ± 0.005	21.7%	0.019–0.025

			2021	3	0.054 ± 0.013	23.9%	0.044–0.068
	Yellow Warbler	*Setophaga petechia*	2007–2019	4	0.021 ± 0.008	39.4%	0.013–0.032
Passerellidae (New World Sparrows)	Green-backed Sparrow	*Arremonops chloronotus*	2007–2019	3	0.076 ± 0.041	53.9%	0.037–0.119
			2021	14	0.028 ± 0.025	90.7%	0.003–0.093
Picidae (Woodpeckers)	Yucatan Woodpecker	*Melanerpes pygmaeus*	2021	2	0.009 ± 0.004	47.5%	0.006–0.013
Pipridae (Manakins)	Red-capped Manakin	*Ceratopipra mentalis*	2007–2019	12	0.013 ± 0.005	38.3%	0.006–0.024
			2021	4	0.003 ± 0.001	27.5%	0.003–0.005
	White-collared Manakin	*Manacus candei*	2007–2019	5	0.024 ± 0.032	134.1%	0.005–0.08
			2021	5	0.003 ± 0.001	45.7%	0.001–0.004
Polioptilidae (Gnatcatchers)	Long-billed Gnatwren	*Ramphocaenus melanurus*	2007–2019	3	0.077 ± 0.055	71.8%	0.04–0.141
			2021	1	0.016	—	—
Scolopacidae (Sandpipers and Allies)	Spotted Sandpiper	*Actitis macularius*	2021	1	0.741	—	—
Thamnophilidae (Typical Antbirds)	Barred Antshrike	*Thamnophilus doliatus*	2007–2019	1	0.167	—	—
			2021	2	0.032 ± 0.031	96.5%	0.01–0.054
	Dusky Antbird	*Cercomacroides tyrannina*	2007–2019	8	0.046 ± 0.014	29.1%	0.026–0.059
			2021	2	0.054 ± 0.058	107.7%	0.013–0.095
	Great Antshrike	*Taraba major*	2021	1	0.09	—	—
Thraupidae (Tanagers and Allies)	Buff-throated Saltator	*Saltator maximus*	2007–2019	1	0.001	—	—
	Gray-headed Tanager	*Eucometis penicillata*	2007–2019	15	0.251 ± 0.119	47.2%	0.101–0.509
			2021	9	0.154 ± 0.059	38.5%	0.083–0.281
	Morelet's Seedeater	*Sporophila morelleti*	2007–2019	15	0.017 ± 0.021	123.7%	0.006–0.091

	Thick-billed Seed-Finch	*Sporophila funerea*	2007–2019	1	0.007	—	—
Tityridae (Tityras and Allies)	Northern Schiffornis	*Schiffornis veraepacis*	2007–2019	1	0.003	—	—
			2021	1	0.002	—	—
	Rose-throated Becard	*Pachyramphus aglaiae*	2007–2019	1	0.05	—	—
Troglodytidae (Wrens)	Spot-breasted Wren	*Pheugopedius maculipectus*	2007–2019	12	0.108 ± 0.055	51.5%	0.027–0.187
			2021	11	0.053 ± 0.081	152.9%	0.009–0.289
	White-bellied Wren	*Uropsila leucogastra*	2007–2019	2	0.224 ± 0.023	10.3%	0.208–0.24
			2021	2	0.074 ± 0.066	89.0%	0.028–0.121
	White-breasted Wood-Wren	*Henicorhina leucosticta*	2007–2019	7	0.194 ± 0.316	163.2%	0.053–0.909
			2021	1	0.28	—	—
Turdidae (Thrushes and Allies)	Clay-colored Thrush	*Turdus grayi*	2021	13	0.021 ± 0.022	104.3%	0.005–0.079
	Swainson's Thrush	*Catharus ustulatus*	2021	5	0.004 ± 0.002	44.7%	0.002–0.007
	Veery	*Catharus fuscescens*	2021	1	0.025	—	—
	White-throated Thrush	*Turdus assimilis*	2007–2019	1	0.038	—	—
	Wood Thrush	*Hylocichla mustelina*	2007–2019	3	0.084 ± 0.032	37.8%	0.054–0.117
Tyrannidae (Tyrant Flycatchers)	Acadian Flycatcher	*Empidonax virescens*	2007–2019	1	0.147	—	—
			2021	4	0.178 ± 0.036	20.0%	0.136–0.216
	Bright-rumped Attila	*Attila spadiceus*	2007–2019	1	0.107	—	—
			2021	2	0.051 ± 0.037	71.5%	0.025–0.077
	Dusky-capped Flycatcher	*Myiarchus tuberculifer*	2007–2019	1	0.031	—	—
			2021	2	0.039 ± 0	1.2%	0.038–0.039
	Eastern Wood-Pewee	*Contopus virens*	2021	1	0.484	—	—

	Eye-ringed Flatbill	*Rhynchocyclus brevirostris*	2007–2019	2	0.066 ± 0.042	64.3%	0.036–0.096
	Great Kiskadee	*Pitangus sulphuratus*	2021	2	0.088 ± 0.001	0.9%	0.087–0.088
	Greenish Elaenia	*Myiopagis viridicata*	2007–2019	1	0.02	—	—
	Northern Bentbill	*Oncostoma cinereigulare*	2007–2019	2	0.107 ± 0.009	8.3%	0.1–0.113
			2021	3	0.044 ± 0.002	4.5%	0.041–0.045
	Ochre-bellied Flycatcher	*Mionectes oleagineus*	2007–2019	28	0.031 ± 0.05	162.8%	0.009–0.278
	Social Flycatcher	*Myiozetetes similis*	2007–2019	1	0.017	—	—
	Sulphur-bellied Flycatcher	*Myiodynastes luteiventris*	2007–2019	2	0.057 ± 0.023	39.7%	0.041–0.073
	Tropical Kingbird	*Tyrannus melancholicus*	2007–2019	1	0.042	—	—
	Tropical Pewee	*Contopus cinereus*	2007–2019	3	0.174 ± 0.042	23.96%	0.126–0.203
	Yellow-bellied Elaenia	*Elaenia flavogaster*	2007–2019	3	0.007 ± 0.003	33.9%	0.005–0.01
	Yellow-bellied Flycatcher	*Empidonax flaviventris*	2007–2019	1	0.101	—	—
			2021	1	0.126	—	—
	Yellow-olive Flycatcher	*Tolmomyias sulphurescens*	2007–2019	1	0.066	—	—
Vireonidae (Vireos, Shrike-Babblers, and Erpornis)	Lesser Greenlet	*Pachysylvia decurtata*	2021	1	0.084	—	—
	Mangrove Vireo	*Vireo pallens*	2007–2019	7	0.173 ± 0.107	61.7%	0.073–0.315
			2021	1	0.019	—	—
	Red-eyed Vireo	*Vireo olivaceus*	2007–2019	2	0.025 ± 0.0	0.0%	0.025–0.025
	White-eyed Vireo	*Vireo griseus*	2007–2019	3	0.057 ± 0.038	66.8%	0.018–0.093
	Yellow-green Vireo	*Vireo flavoviridis*	2007–2019	13	0.02 ± 0.008	40.3%	0.009–0.035
			2021	1	0.007	—	—

Appendix 3. Summary of 75 abundant, widespread species with relatively high trophic positions that could be useful sentinels for assessing avian MeHg exposure in various Neotropical habitats. An asterisks (*) indicates that other members within this group may also be useful. Relative abundance was evaluated by visually inspecting mist-netting capture data and country-level eBird Bar Charts (https://ebird.org/GuideMe?cmd=changeLocation). The exclusion of a family or species from this list does not necessarily signify that those taxa would be poor MeHg bioindicators, but that sample acquisition could be challenging. Focal species selection for a given research effort should ultimately be informed by their abundance at the intended sampling locations, their diet and trophic niche, as well as the geographic scale of interest. Species are arranged alphabetically by order and family. Species highlighted in green may be especially useful due to their widespread range.

Order	Family	Common name	Latin name	Trophic niche	Migratory status	Primary habitat(s)	Region(s) occupied
Caprimulgiformes	**Caprimulgidae (Nightjars and Allies) ***	Common Pauraque	*Nyctidromus albicollis*	Invertivore	Resident	Tropical lowland evergreen forest; secondary forest; tropical deciduous forest	Texas, USA to Argentina
	Trochilidae (Hummingbirds) *	Rufous-tailed Hummingbird	*Amazilia tzacatl*	Nectarivore	Resident	Tropical lowland evergreen forest; secondary forest	Mexico to Ecuador
		White-necked Jacobin	*Florisuga mellivora*	Nectarivore	Resident	Tropical lowland evergreen forest; secondary forest	Mexico to Bolivia
	(Sandpipers and Allies) *	Spotted Sandpiper	*Actitis macularius*	Aquatic predator	Full migrant	Riverine sand beaches; freshwater lakes and ponds	Canada to Argentina; West Indies
Coraciiformes *	**Alcedinidae (Kingfishers) ***	American Pygmy Kingfisher	*Chloroceryle aenea*	Aquatic predator	Resident	Streams; freshwater lakes and ponds	Mexico to Paraguay
		Green Kingfisher	*Chloroceryle americana*	Aquatic predator	Resident	Streams; freshwater lakes and ponds	Texas, USA to Argentina
	Momotidae (Motmots) *	Lesson's Motmot	*Momotus lessonii*	Omnivore	Resident	Tropical lowland evergreen forest; montane evergreen forest; secondary	Mexico to Panama

						forest	
Cuculiformes *	**Cuculidae (Cuckoos) ***	Groove-billed Ani	*Crotophaga sulcirostris*	Invertivore	Resident	Second-growth scrub; riparian thickets	Texas, USA to Peru
		Squirrel Cuckoo	*Piaya cayana*	Invertivore	Resident	Tropical lowland evergreen forest; tropical deciduous forest; secondary forest	Mexico to Uruguay
Galbuliformes	**Galbulidae (Jacamars) ***	Rufous-tailed Jacamar	*Galbula ruficauda*	Invertivore	Resident	Tropical lowland evergreen forest; gallery forest	Mexico to Brazil
Passeriformes	**Cardinalidae (Cardinals and Allies) ***	Hepatic Tanager	*Piranga flava*	Invertivore	Resident	Pine-oak forest; gallery forest; tropical deciduous forest	Colorado, USA to Argentina
		Red-crowned Ant-Tanager	*Habia rubica*	Invertivore	Resident	Tropical lowland evergreen forest	Mexico to Brazil
		Red-throated Ant-Tanager	*Habia fuscicauda*	Invertivore	Resident	Tropical lowland evergreen forest; flooded tropical evergreen forest	Mexico to Colombia
		Summer Tanager	*Piranga rubra*	Invertivore	Partial migrant	Gallery forest	Iowa, USA to Bolivia; West Indies
	Formicariidae (Antthrushes) *	Mayan Antthrush	*Formicarius moniliger*	Invertivore	Resident	Tropical lowland evergreen forest; flooded tropical evergreen forest	Mexico to Honduras
	Furnariidae (Ovenbirds and Woodcreepers) *	Ivory-billed Woodcreeper	*Xiphorhynchus flavigaster*	Invertivore	Resident	Tropical lowland evergreen forest; montane evergreen forest; tropical deciduous forest	Mexico to Costa Rica
		Olivaceous Woodcreeper	*Sittasomus griseicapillus*	Invertivore	Resident	Tropical lowland evergreen	Mexico to Uruguay

					forest; montane evergreen forest; tropical deciduous forest	
	Plain Xenops	*Xenops minutus*	Invertivore	Resident	Tropical lowland evergreen forest; flooded tropical evergreen forest	Mexico to Brazil
	Ruddy Woodcreeper	*Dendrocincla homochroa*	Invertivore	Resident	Tropical lowland evergreen forest; montane evergreen forest; tropical deciduous forest	Mexico to Venezuela
	Tawny-winged Woodcreeper	*Dendrocincla anabatina*	Invertivore	Resident	Tropical lowland evergreen forest	Mexico to Panama
	Wedge-billed Woodcreeper	*Glyphorynchus spirurus*	Invertivore	Resident	Tropical lowland evergreen forest; montane evergreen forest	Mexico to Brazil
Hirundinidae (Swallows) *	Barn Swallow	*Hirundo rustica*	Invertivore	Partial migrant	Pastures/agricultural lands; Northern temperate grassland	Worldwide
	Gray-breasted Martin	*Progne chalybea*	Invertivore	Partial migrant	Second-growth scrub; pastures/agricultural lands	Mexico to Argentina
	Northern Rough-winged Swallow	*Stelgidopteryx serripennis*	Invertivore	Partial migrant	Second-growth scrub; pastures/agricultural lands	Canada to Panama; West Indies
Oxyruncidae (Sharpbill, Royal Flycatcher, and Allies) *	Royal Flycatcher	*Onychorhynchus coronatus*	Invertivore	Resident	Tropical lowland evergreen forest	Mexico to Brazil
Parulidae (New World Warblers) *	American Redstart	*Setophaga ruticilla*	Invertivore	Full migrant	Tropical lowland evergreen forest; montane evergreen forest	Canada to Peru; West Indies

	Black-and-white Warbler	*Mniotilta varia*	Invertivore	Full migrant	Tropical lowland evergreen forest; montane evergreen forest	Canada to Ecuador; West Indies
	Common Yellowthroat	*Geothlypis trichas*	Invertivore	Partial migrant	Freshwater/saltwater/brackish marshes	Canada to Panama; West Indies
	Golden-crowned Warbler	*Basileuterus culicivorus*	Invertivore	Resident	Tropical lowland evergreen forest; montane evergreen forest	Mexico to Argentina
	Gray-crowned Yellowthroat	*Geothlypis poliocephala*	Invertivore	Resident	Second-growth scrub; riparian thickets	Mexico to Panama
	Hooded Warbler	*Setophaga citrina*	Invertivore	Full migrant	Tropical lowland evergreen forest; secondary forest	Canada to Venezuela; West Indies
	Northern Waterthrush	*Parkesia noveboracensis*	Invertivore	Full migrant	Tropical lowland evergreen forest; secondary forest	Alaska, USA to Ecuador; West Indies
	Yellow Warbler	*Setophaga petechia*	Invertivore	Partial migrant	Gallery forest; secondary forest; mangrove forest	Alaska, USA to Peru; West Indies
Passerellidae (New World Sparrows) *	Green-backed Sparrow	*Arremonops chloronotus*	Omnivore	Resident	Tropical lowland evergreen forest; tropical deciduous forest	Mexico to Honduras
	Orange-billed Sparrow	*Arremon aurantiirostris*	Omnivore	Resident	Tropical lowland evergreen forest	Mexico to Peru
Polioptilidae (Gnatcatchers) *	Blue-gray Gnatcatcher	*Polioptila caerulea*	Invertivore	Partial migrant	Tropical deciduous forest; tropical lowland evergreen forest	Canada to Honduras; West Indies
	Tropical Gnatcatcher	*Polioptila plumbea*	Invertivore	Resident	Tropical deciduous	Mexico to Brazil

					forest; tropical lowland evergreen forest; Arid lowland/montane scrub	
Thamnophilidae (Typical Antbirds) *	Barred Antshrike	*Thamnophilus doliatus*	Invertivore	Resident	Second-growth scrub; riparian thickets; river island scrub	Mexico to Argentina
	Dot-winged Antwren	*Microrhopias quixensis*	Invertivore	Resident	Tropical lowland evergreen forest	Mexico to Brazil
	Dusky Antbird	*Cercomacroides tyrannina*	Invertivore	Resident	Tropical lowland evergreen forest; secondary forest	Mexico to Brazil
	Great Antshrike	*Taraba major*	Invertivore	Resident	Tropical lowland evergreen forest; secondary forest; riparian thickets	Mexico to Argentina
Thraupidae (Tanagers and Allies) *	Gray-headed Tanager	*Eucometis penicillata*	Invertivore	Resident	Tropical lowland evergreen forest; gallery forest; secondary forest	Mexico to Paraguay
Troglodytidae (Wrens) *	Gray-breasted Wood-Wren	*Henicorhina leucophrys*	Invertivore	Resident	Montane evergreen forest	Mexico to Bolivia
	House Wren	*Troglodytes aedon*	Invertivore	Resident	Second-growth scrub; arid lowland/montane scrub	Canada to Chile
	Spot-breasted Wren	*Pheugopedius maculipectus*	Invertivore	Resident	Tropical lowland evergreen forest; secondary forest	Mexico to Costa Rica
	White-breasted Wood-Wren	*Henicorhina leucosticta*	Invertivore	Resident	Tropical lowland evergreen forest; montane evergreen forest	Mexico to Peru

Turdidae (Thrushes and Allies) *	Clay-colored Thrush	*Turdus grayi*	Omnivore	Resident	Tropical lowland evergreen forest; tropical deciduous forest; secondary forest	Texas, USA to Colombia
	Swainson's Thrush	*Catharus ustulatus*	Invertivore	Full migrant	Montane evergreen forest; tropical lowland evergreen forest	Alaska, USA to Argentina
	Wood Thrush	*Hylocichla mustelina*	Invertivore	Full migrant	Tropical lowland evergreen forest; secondary forest	Canada to Colombia
Tyrannidae (Tyrant Flycatchers) *	Boat-billed Flycatcher	*Megarynchus pitangua*	Invertivore	Resident	Tropical lowland evergreen forest; secondary forest	Mexico to Argentina
	Bright-rumped Attila	*Attila spadiceus*	Invertivore	Resident	Tropical lowland evergreen forest; secondary forest; montane evergreen forest	Mexico to Bolivia
	Common Tody-Flycatcher	*Todirostrum cinereum*	Invertivore	Resident	Tropical lowland evergreen forest; montane evergreen forest; tropical deciduous forest	Mexico to Brazil
	Dusky-capped Flycatcher	*Myiarchus tuberculifer*	Invertivore	Resident	Montane evergreen forest; tropical lowland evergreen forest; tropical deciduous forest	Arizona, USA to Argentina
	Eastern Wood-Pewee	*Contopus virens*	Invertivore	Full migrant	Tropical lowland evergreen forest; secondary forest	Canada to Peru; West Indies

	Great Kiskadee	*Pitangus sulphuratus*	Omnivore	Resident	Secondary forest; riparian thickets	Texas, USA to Argentina
	Sepia-capped Flycatcher	*Leptopogon amaurocephalus*	Invertivore	Resident	Tropical lowland evergreen forest; secondary forest	Mexico to Brazil
	Social Flycatcher	*Myiozetetes similis*	Invertivore	Resident	Tropical lowland evergreen forest; tropical deciduous forest	Mexico to Argentina
	Tropical Kingbird	*Tyrannus melancholicus*	Invertivore	Resident	Secondary forest; gallery forest; river-edge forest	Arizona, USA to Argentina
	Tropical Pewee	*Contopus cinereus*	Invertivore	Resident	Tropical lowland evergreen forest; montane evergreen forest	Mexico to Argentina
	Yellow-bellied Elaenia	*Elaenia flavogaster*	Invertivore	Resident	Second-growth scrub; riparian thickets	Mexico to Brazil
	Yellow-olive Flycatcher	*Tolmomyias sulphurescens*	Invertivore	Resident	Tropical lowland evergreen forest; montane evergreen forest; tropical deciduous forest	Mexico to Uruguay
Vireonidae (Vireos, Shrike-Babblers, and Erpornis) *	Lesser Greenlet	*Pachysylvia decurtata*	Invertivore	Resident	Tropical lowland evergreen forest; tropical deciduous forest	Mexico to Ecuador
	Mangrove Vireo	*Vireo pallens*	Invertivore	Resident	Tropical deciduous forest; mangrove forest	Mexico to Costa Rica
	Red-eyed Vireo	*Vireo olivaceus*	Invertivore	Full migrant	Tropical lowland evergreen forest; secondary forest	Canada to Bolivia

		Rufous-browed Peppershrike	*Cyclarhis gujanensis*	Invertivore	Resident	Tropical lowland evergreen forest; gallery forest	Mexico to Argentina
		Tawny-crowned Greenlet	*Tunchiornis ochraceiceps*	Invertivore	Resident	Tropical lowland evergreen forest	Mexico to Bolivia
Pelecaniformes	**Ardeidae (Herons, Egrets, and Bitterns) ***	Black-crowned Night-Heron	*Nycticorax nycticorax*	Aquatic predator	Partial migrant	Freshwater/saltwater/brackish marshes; freshwater lakes and ponds; rivers	Worldwide
		Great Egret	*Ardea alba*	Aquatic predator	Partial migrant	Freshwater/saltwater/brackish marshes; coastal sand beaches/mudflats	Worldwide
		Green Heron	*Butorides virescens*	Aquatic predator	Partial migrant	Freshwater lakes, ponds, and marshes	United States to Ecuador; West Indies
	Pelecanidae (Pelicans) *	Brown Pelican	*Pelecanus occidentalis*	Aquatic predator	Partial migrant	Coastal waters	United States to Peru; West Indies
Podicipediformes *	**Podicipedidae (Grebes) ***	Least Grebe	*Tachybaptus dominicus*	Aquatic predator	Resident	Freshwater marshes, lakes, and ponds	Texas, USA to Uruguay
		Pied-billed Grebe	*Podilymbus podiceps*	Aquatic predator	Resident	Freshwater lakes, ponds, and marshes	Canada to Argentina; West Indies
Suliformes	**Anhingidae (Anhingas)**	Anhinga	*Anhinga anhinga*	Aquatic predator	Resident	Freshwater lakes and ponds; rivers	United States to Uruguay
	Fregatidae (Frigatebirds)	Magnificent Frigatebird	*Fregata magnificens*	Aquatic predator	Resident	Coastal waters; pelagic waters; mangrove forest	United States to Uruguay; Cape Verde
	Phalacrocoracidae (Cormorants and Shags) *	Neotropic Cormorant	*Phalacrocorax brasilianus*	Aquatic predator	Resident	Coastal waters; freshwater lakes and ponds; rivers	Texas, USA to Chile; Cuba
	Sulidae (Boobies and Gannets) *	Brown Booby	*Sula leucogaster*	Aquatic predator	Resident	Coastal waters; pelagic waters	Worldwide