Environmental Toxicology and Chemistry—Volume 41, Number 10—pp. 2479–2487, 2022
Received: 1 January 2022 | Revised: 31 January 2022 | Accepted: 19 July 2022

Environmental Toxicology

Bioaccumulation of Mercury and Radiocesium in Waterfowl Introduced to a Site with Legacy Contamination

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Abstract: Despite the propensity of waterfowl species to readily accumulate anthropogenic contaminants within polluted environments, few studies have examined bioaccumulation rates over time when entering such a contaminated site. We examined mercury (Hg) and radiocesium (^{137}Cs) bioaccumulation over time in two waterfowl species released into a wetland system containing legacy contamination on the US Department of Energy's Savannah River Site in South Carolina. Released birds were collected at select time intervals over an exposure period of 94 days. We quantified total Hg concentrations in blood, muscle, and liver tissues, and ^{137}Cs activity in whole-body and muscle tissues. The relationship between the contaminant burdens of different body tissue types was examined over time. Likely a result of microhabitat selection, mallards in our study readily accumulated both Hg and ^{137}Cs at consistent rates over time within our study system, while ring-neck ducks did not. The findings demonstrated that whole blood can be used as a robust, nondestructive sampling alternative to estimate Hg burdens within muscle and liver, and whole-body ^{137}Cs activity is a good predictor of muscle burdens. Understanding such bioaccumulation information in waterfowl is useful for the assessment of the potential health risk in wildlife, as well as being important for human risk assessment toward the consumption of popular game species. *Environ Toxicol Chem* 2022;41:2479–2487. © 2022 The Authors. *Environmental Toxicology and Chemistry* published by Wiley Periodicals LLC on behalf of SETAC.

Keywords: Bioaccumulation; Mallard; Mercury; Radiocesium; Waterfowl; Wildlife toxicology

INTRODUCTION

Anthropogenic and natural pollution into the environment has been of great concern for wildlife species worldwide (Driscoll et al., 2013; Isaksson, 2010). Contaminants such as trace elements and radionuclides have the propensity to readily bioaccumulate in animal tissues through dietary exposure (Dietz et al., 2000; Evers, 2018). Although historically more attention has focused on monitoring for the presence of anthropogenic contaminants in higher trophic level fish and wildlife, there is growing interest in assessing contaminant exposures in wildlife more broadly, including game species commonly consumed by hunters and their families (Cristol et al., 2012; Oldenkamp et al., 2017). Waterfowl are of particular concern given their mobility (i.e., large home ranges and migratory paths that can extend thousands of kilometers from a contaminant source; Kennamer, 2003) and they can provide a potential avenue for contaminants to enter the human food web far from point sources (Conder & Arblaster, 2016; Cristol et al., 2012; Oldenkamp et al., 2017).

Once absorbed into the body in liver, kidneys, and the nervous system, elevated concentrations of certain trace elements, such as mercury (both inorganic [Hg] and organic [MeHg]), can induce toxicological effects in birds (Chételat et al., 2020; Scheuhammer et al., 2007; Whitney & Cristol, 2018). Higher MeHg exposure may also be associated with increased infection and disease in birds (Scheuhammer et al., 2007). Reproduction is also negatively affected in birds exposed to MeHg (Chételat et al., 2020; Fuchsman et al., 2017; Whitney & Cristol, 2018), with maternal MeHg burdens deposited into fertile eggs (Ackerman et al., 2016). At concentrations exceeding species-specific thresholds, embryos may exhibit development abnormalities and an increased prenatal morality (Heinz et al., 2009).

Exposure to radionuclides, such as radiocesium (^{137}Cs), may also lead to various health problems, including genetic damage. Ellegren et al. (1997) found DNA mutation rates in

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Published online 22 July 2022 in Wiley Online Library (wileyonlinelibrary.com).
DOI: 10.1002/etc.5444

barn swallows from the Chernobyl disaster area were up to tenfold higher than those from reference sites, expressing albinism among those affected and potentially causing population declines as a result of loss of fitness among individuals. Furthermore, birds exposed to radionuclides in the Chernobyl Exclusion Zone were also suggested to have an increased rate of tumor formations (Møller et al., 2013). Although waterfowl exposed to ^{137}Cs in less contaminated environments, such as the US Department of Energy's Savannah River Site, do not exhibit activity concentrations high enough to increase mutation rates (George et al., 1991; Johnson et al., 1999), birds can contain ^{137}Cs levels higher than that advised by the European Economic Community (EEC, 1986) guidelines for safe consumption by humans (Kennamer et al., 2017). Given that ^{137}Cs is a chemical analog for potassium (Caldwell et al., 2011) that readily accumulates in the edible skeletal muscle of exposed organisms, humans can potentially be exposed to ^{137}Cs through the consumption of contaminated meat (Kennamer et al., 2017; Oldenkamp et al., 2017).

Although it is well established that both Hg and ^{137}Cs can accumulate in exposed waterfowl, knowledge of the rate of accumulation over time for these contaminants within the environment is lacking, with few aviary studies in existence for birds under laboratory conditions for Hg (Bearhop et al., 2000; Heinz, 1987). Leaphart et al. (2020) modeled arsenic and selenium bioaccumulation over time in ring-necked ducks experimentally restricted to a basin containing coal fly ash, although Hg bioaccumulation was negligible in that study, likely given its bioavailability and presence of selenium. Studies conducted on the US Department of Energy's Savannah River Site have examined accumulation rates of ^{137}Cs in wood ducks (Fendley et al., 1977) and ring-necked ducks (Kennamer et al., 2017), but did not compare potential effects of differences in foraging strategies (i.e., collection of dietary items via dabbling vs. diving). The goal of the present study was to quantify bioaccumulation of Hg and ^{137}Cs in waterfowl predominately using a contaminated wetland. Our objectives were to (1) examine the bioaccumulation of total Hg and ^{137}Cs in mallards (*Anas platyrhynchos*) and ring-necked ducks (*Aythya collaris*) exposed over time, (2) compare accumulation rates over time between these two species with differing foraging strategies (foraging for food via dabbling vs. diving), and (3) develop predictive models to assess the utility of nondestructive sampling methods for estimating exposure in muscle (Hg and ^{137}Cs) and liver (Hg only) to these contaminants. If ^{137}Cs and Hg bioaccumulate within both species examined in our study, we predicted that mallards would accumulate greater contaminant burdens at a faster rate compared to ring-necked ducks given their closer association with contaminated sediments within the study system, and that nondestructive sampling measures would be effective for estimating ^{137}Cs activity and Hg concentrations in other organs.

METHODS

Study area

Our study was conducted on a Hg and ^{137}Cs contaminated wetland on the US Department of Energy's Savannah River Site located near Aiken, South Carolina. A former nuclear production and research facility, the 780 km^2 Savannah River Site landscape is now a National Environmental Research Park (White & Gaines, 2000) comprising managed pine forests, bottomland hardwoods, and various wetlands and waterbodies (Kilgo & Blake, 2005). Many of the wetlands and reservoirs on the Savannah River Site act as wintering refuge and stopover habitat for up to 28 migrating waterfowl species (Kennamer, 2005), many of which accumulate contaminants at concentrations higher than US Environmental Protection Agency (USEPA) and EEC recommended guidelines for human consumption of contaminated meat (Kennamer et al., 2017; Oldenkamp et al., 2017). Of those aquatic systems, Fourmile Branch is used by several species of migratory and resident waterfowl, including overwintering mallard (*Anas platyrhynchos*) and ring-necked ducks (*Aythya collaris*). Located approximately 1.5 km southwest of the Savannah River Site H-Area, a storage facility for liquid waste material, the Fourmile Beaver Pond has historically received direct input of anthropogenic waste from an input canal, including both ^{137}Cs (Carlton et al., 1992; Haselow et al., 1990) and Hg (Kvartek et al., 1994; Xu et al., 2019). From 1955 to 1988, it is suspected that there were occasional chemical releases from H-Area through the bottom of seepage basins due to the shallow depth of ground water (~6 m; Carlton et al., 1992; Kvartek et al., 1994; Xu et al., 2019) upstream from this wetland. Fourmile Beaver Pond is made up of two microhabitats: a main lentic system pond characterized by a beaver dam separating the shallow pond from a branch and downstream riparian wetland. Although data on ^{137}Cs activity and Hg concentrations in waterfowl and their dietary items are lacking for this specific study site, studies have shown elevated Hg concentrations in sediment media upstream of the pond from the seepage basin into the Fourmile Branch system (Xu et al., 2019).

Bird processing and collection

For our study, we purchased 48 farm-raised mallard drakes in December 2015 and January 2016 to be released onto the Fourmile Beaver Pond system to study Hg and ^{137}Cs accumulation. Male ring-necked ducks ($n = 30$) were also live-trapped from a reservoir on the Savannah River Site (L-lake), where contamination levels have been previously observed to be relatively low in this species (Kennamer et al., 2017; Oldenkamp et al., 2017). Prior to release, all ducks were weighed and banded with a numbered aluminum leg band and colored nasal saddle for identification. Blood samples (1.0 ml) were also collected from the brachial vein of each duck to assess initial circulating Hg concentrations within the blood, and a subset of random individuals were whole-body counted (method description in Radiocesium Counting Procedures) for 15-min live-count intervals to determine pre-existing ^{137}Cs activity. All ducks were rendered flightless by wing clipping the primary flight feathers on one wing and released onto Fourmile Beaver Pond within 24 h. Mallards ($n = 27$) and ring-necked ducks ($n = 17$) were subsequently collected using shotguns at known time intervals ranging from 4 to 94 days of exposure to

determine bioaccumulation rates for both Hg and ^{137}Cs. Blood samples (≥1.0 ml) were collected via cardiac puncture and dead birds were transported to the Savannah River Ecology Laboratory to be reweighed and frozen at −20 °C until later processing. Subsequently, whole-body ^{137}Cs burdens were determined and birds were then dissected to obtain muscle and liver samples that could be analyzed for Hg and ^{137}Cs. All animal handling practices and euthanasia were performed with accompanying state and federal collection permits, and in accordance with University of Georgia Institutional Animal Care and Use guidelines under protocol A2012 12-010-Y3-A5.

Radiocesium counting procedures

Whole-body ^{137}Cs burdens were determined in a subset of birds prior to release and all birds after collection from Fourmile. All birds were packed into a Lucite cylinder and analyzed using Canberra Genie (2000) gamma spectroscopy, centered on 662 kiloelectron-volts (keV) with a region of interest of 596–728 keV to record total ^{137}Cs emissions. Radiocesium was detected using a NaI (Tl) gamma detector (10.2 cm × 15.2 cm; Bicron Model 6H3Q/5) contained in a lead brick counting chamber (81 × 81 × 107 cm). The system was manually calibrated daily using a traceable ^{137}Cs source (Catalogue No. NES-101S; New England Nuclear Gamma Reference Disc Source Set). Whole frozen birds and background samples were counted for 30 min. Whole-body ^{137}Cs activity (Becquerels [Bq], Bq/g, fresh whole-body mass) were calculated using background-corrected count rates derived from background and aqueous standards with known ^{137}Cs quantities and mass-specific count yield estimates as described in Kennamer et al. (2017).

Freeze-dried, homogenized muscle samples were subsampled, weighed, and analyzed for ^{137}Cs. Samples were analyzed using a Packard Cobra (Auto-Gamma Counter, Model Cobra II 5003; Packard Instruments) with a single 3-in. through-hole NaI detector to measure ^{137}Cs emissions at 662 keV photons between a region of interest of 580–754 keV. Samples were counted for 60-min intervals with empty scintillation vials (serving as background samples) arranged in every fifth counting position. Prior to sample analysis, four standards of differing weight (1-g increments from 1 to 4 g of dry mass) were produced by combining commercially purchased chicken breast and known quantities of ^{137}Cs (745 Bq in each sample, decay-corrected for the date of preparation) to calculate an average count yield value (0.2213). We then divided our background-adjusted dry tissue count rates by the averaged count yield to determine ^{137}Cs activity in mallard muscle samples (in Bq, Bq/g [dry mass]). Minimum detectable concentrations were calculated as described by Currie (1968).

Hg analysis procedure

Homogenized muscle, liver, and blood samples (pre- and postexposure) from each bird were also analyzed for total Hg (THg) in accordance to USEPA method 7473 (USEPA, 2007). Approximately 30–50-mg subsamples of freeze-dried tissue were analyzed by thermal decomposition, catalytic conversion, amalgamation, and atomic absorption spectrophotometry (DMA-80; Milestone), with an instrument detection limit of 0.01 ng of THg. For quality assurance, a replicate, sample blanks, and two certified reference materials (CRMs, TORT-3 lobster hepatopancreas and PACS-2 marine sediment; National Research Council of Canada) were analyzed for each set of 10 samples, and solid CRMs were used to calibrate the instrument. Method detection limits (threefold the standard deviation of procedural blanks) averaged 51.6 ng/kg dry mass for muscle, liver, and blood samples. Mean percent recoveries of THg for the CRMs averaged 99.2% (range 88.3%–110.5%, $n = 46$), where TORT-3 and PACS-2 were 102.7% (range 96.5%–110.5%, $n = 23$) and 95.6% (range 88.3%–104.6%, $n = 23$), respectively. Mercury concentrations for our samples are presented as mg/kg Hg (mass equivalent to parts per million [ppm]) on a dry mass basis and are representative of THg. Mercury speciation was not considered in the present study, although it is known that >80% of Hg in biological samples collected from waterfowl is the methylmercury (MeHg; Sullivan & Kopec, 2018).

Statistical analyses

Before modeling contaminant uptake over time, we calculated the change in the percentage of body mass of each bird between initial handling and final harvest to ensure Hg and ^{137}Cs accumulation was not influenced by a lack of food availability or adequate foraging while restricted to the study area. We then used linear regression models to examine weight change over the time that individuals resided at the Fourmile Beaver Pond study area.

To ensure ducks sampled from our control area or purchased from commercial suppliers did not enter the study with elevated prior Hg exposure, we examined pre-exposure THg blood levels for all individuals. Any individuals with blood THg levels greater than three standard deviations above the mean for that species were excluded from any analyses. For ^{137}Cs, we observed concentrations below background levels, resulting in negative values. Despite this, we included all negative values and concentrations below their minimum detectable concentrations in our analyses to avoid any bias of our overall results associated with not reporting such values or replacing them with an arbitrary indicator value (Gilbert & Kinnison, 1981; Newman et al., 1989).

All data were then tested for normality and homoscedasticity using the Shapiro–Wilk test and nonconstant variance score test, respectively, using R Studio statistical software (package stats, Ver 3.3.1; R Development Core Team). Contaminant data were then natural log-transformed, when appropriate, to approximate normal distributions and homoscedasticity; specific transformations are detailed for individual analyses in the results. We then examined bioaccumulation of Hg over time using separate nonlinear third-degree polynomial regression models for blood, muscle, and liver. We used linear regression models to examine the relationship between Hg concentrations in blood, liver, and muscle.

To model ^{137}Cs bioaccumulation, we used the classic Richards model modified for contaminant uptake (Leaphart et al., 2019; Peters & Brisbin, 1996; Potter, 1987). This model

provides information on the maximum bioaccumulation of ^{137}Cs in ducks utilizing the Fourmile Beaver Pond system, as well as the amount of time necessary to reach that steady-state equilibrium concentration. The modified Richards model used is defined by the equation

$$C_t = \left[\left(C_e^{(1-m)} - \left(C_e^{(1-m)} - C_0^{(1-m)}\right)\right) \times \exp\left(\frac{-2t}{T}(m+1)\right)\right]^{1/(1-m)}$$

where C_t is the concentration of the contaminant in the organism at time t, C_e is the final contaminant concentration at equilibrium where contaminant uptake is equal to elimination, C_0 is the concentration prior to exposure, t is the exposure period, T is the amount of time for C_t to reach 95% of the maximum concentration at equilibrium, and m is the Richards shape parameter which for our study; we used the constant value 0 to evaluate the classic sigmoidal model (Brisbin et al., 1990; Leaphart et al., 2019; Richards, 1959). Similar to our Hg data, whole-body ^{137}Cs activity was compared to muscle ^{137}Cs using linear regression models.

RESULTS

Of the ducks released into the Fourmile Beaver Pond system, we were able to collect 27 of our released mallards over a 94-day exposure period and 17 ring-necked ducks over 60 days, none of which were excluded from our study due to elevated pre-exposure. Neither mallard nor ring-necked ducks exhibited weight loss over time (mallards, $F_{1,25} = 0.94$, $p = 0.34$; ring-necked ducks, $F_{1,15} = 1.81$, $p = 0.20$), suggesting all birds were able to find adequate food during the study and on average did not lose weight. However, linear regression models for ring-necked ducks showed no Hg bioaccumulation in blood ($R^2 = -0.09$, $F_{3,13} = 0.57$, $p = 0.64$), muscle ($R^2 = -0.08$, $F_{3,13} = 0.63$, $p = 0.61$), or liver ($R^2 = -0.01$, $F_{3,13} = 0.95$, $p = 0.45$) over time. Therefore, Hg concentrations in blood, muscle, and liver remained 0.07 (±0.006 SE), 0.05 (±0.005 SE), and 0.26 (±0.025 SE) mg/kg dry weight, respectively, during the present study. Furthermore, an added Wilcoxon signed-rank test revealed no difference ($p = 0.38$) between pre- and post-exposure whole-body ^{137}Cs activity in ring-necked ducks, and birds had 0.005 (±0.004 SE) Bq/g ^{137}Cs for the entire sampling period. For this reason, no further comparisons were made between the accumulation of Hg and ^{137}Cs, or among tissues, of ring-necked ducks in the present study.

Unlike the ring-necked ducks, mallards readily bioaccumulated contaminants during the 94-day exposure period on Fourmile Beaver Pond. Not only was Hg accumulation in blood positively associated with exposure time ($R^2 = 0.87$, $F_{3,23} = 59.44$, $p < 0.001$), Hg also readily bioaccumulated in both muscle ($R^2 = 0.87$, $F_{3,23} = 61.55$, $p < 0.001$) and liver ($R^2 = 0.90$, $F_{3,23} = 82.76$, $p < 0.001$) as well (Figure 1). Whole-body radiocesium activity was estimated to reach maximum activity of 0.15 Bq/g ^{137}Cs (wet wt, t-value = 17.18, SE = 0.009)

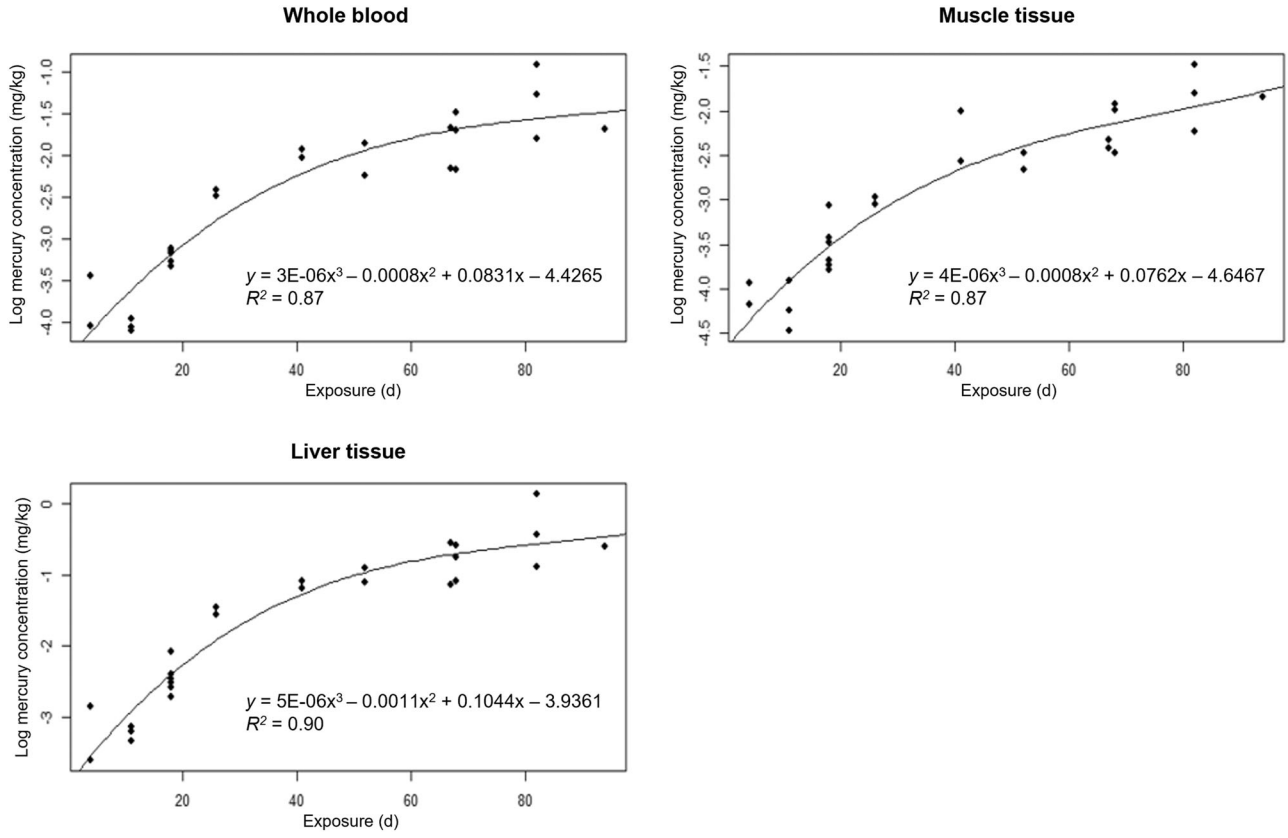

FIGURE 1: Total mercury bioaccumulation in blood, muscle, and liver of mallard ducks (*Anas platyrhynchos*) released on a contaminated wetland for known exposure periods on the US Department of Energy's Savannah River Site in Aiken, South Carolina.

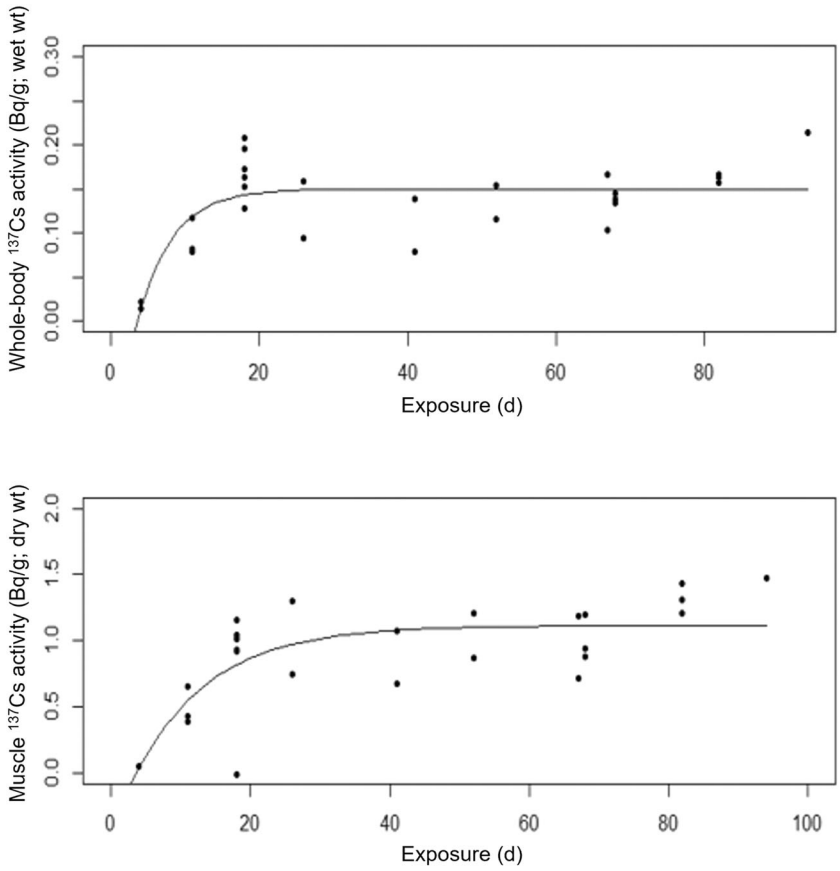

FIGURE 2: Bioaccumulation of radiocesium (^{137}Cs) by mallard ducks (*Anas platyrhynchos*) released on a contaminated wetlands for known exposure periods on the US Department of Energy's Savannah River Site in Aiken, South Carolina.

after 9.44 days (t-value = 2.25, SE = 4.200) of exposure (Figure 2). Much of this ^{137}Cs was in muscle, which reached a maximum activity of 1.11 Bq/g ^{137}Cs (dry wt, t-value = 13.34, SE = 0.08) after an estimated 22 days (t-value = 3.0, SE = 7.30) of exposure (Figure 2).

When examining the relationship between Hg concentrations and ^{137}Cs activity in different tissues, we found that all sample-type comparisons presented significant, positive relationships within respective contaminants (Figure 3). Log-transformed linear modeling revealed blood can be used as an effective measure to estimate Hg concentrations in both muscle ($R^2 = 0.95$, $F_{1,25} = 438.1$, $p < 0.001$) and liver ($R^2 = 0.98$, $F_{1,25} = 1038$, $p < 0.001$). Similarly, either muscle or liver could be used to effectively estimate the other given their positive relationship of Hg concentrations ($R^2 = 0.95$, $F_{1,25} = 503.1$, $p < 0.001$). Although not as precise, linear modeling of whole-body radiocesium activity concentrations from live birds also was an effective estimator of freeze-dried muscle tissues ($R^2 = 0.52$, $F_{1,25} = 27.39$, $p < 0.001$).

DISCUSSION

For many decades, waterfowl have been shown to be important accumulators of various anthropogenic pollutants, including heavy metals and radionuclides (Cristol et al., 2012; Fendley et al., 1977; Fimreite et al., 1971; Golden & Rattner, 2003; Hernández et al., 1999; Leaphart et al., 2020). Given their mobility and propensity to use multiple waterbodies across large spatial scales, data on time-specific bioaccumulation rates of contaminants are needed to better understand potential risks to waterfowl as well as to humans and other predators (Leaphart et al., 2020). Our data demonstrate that ring-necked ducks accumulated neither Hg nor ^{137}Cs during the present study period, whereas mallards readily accumulated both contaminants in blood, muscle, and liver. This indicates that at the contaminated study site, mallards are a better sentinel for biomonitoring compared to diving duck species, such as the ring-necked duck. Furthermore, our data suggest a strong relationship between Hg concentrations in blood, muscle, and liver in mallards, suggesting whole blood can reliably be used as a nondestructive sampling technique to estimate Hg concentrations in waterfowl tissues. Similarly, nondestructive sampling could be used to assess ^{137}Cs activity, given our observed relationship between muscle tissue and whole-body ^{137}Cs activity.

Contaminants in many landscapes are heterogeneously distributed, and an individual's movement and resource selection within habitats can influence contaminant exposure and accumulation rates over time (Hinton et al., 2019; Kearns et al., 2019). Indeed, microhabitat selection associated with foraging preference and free movement throughout the

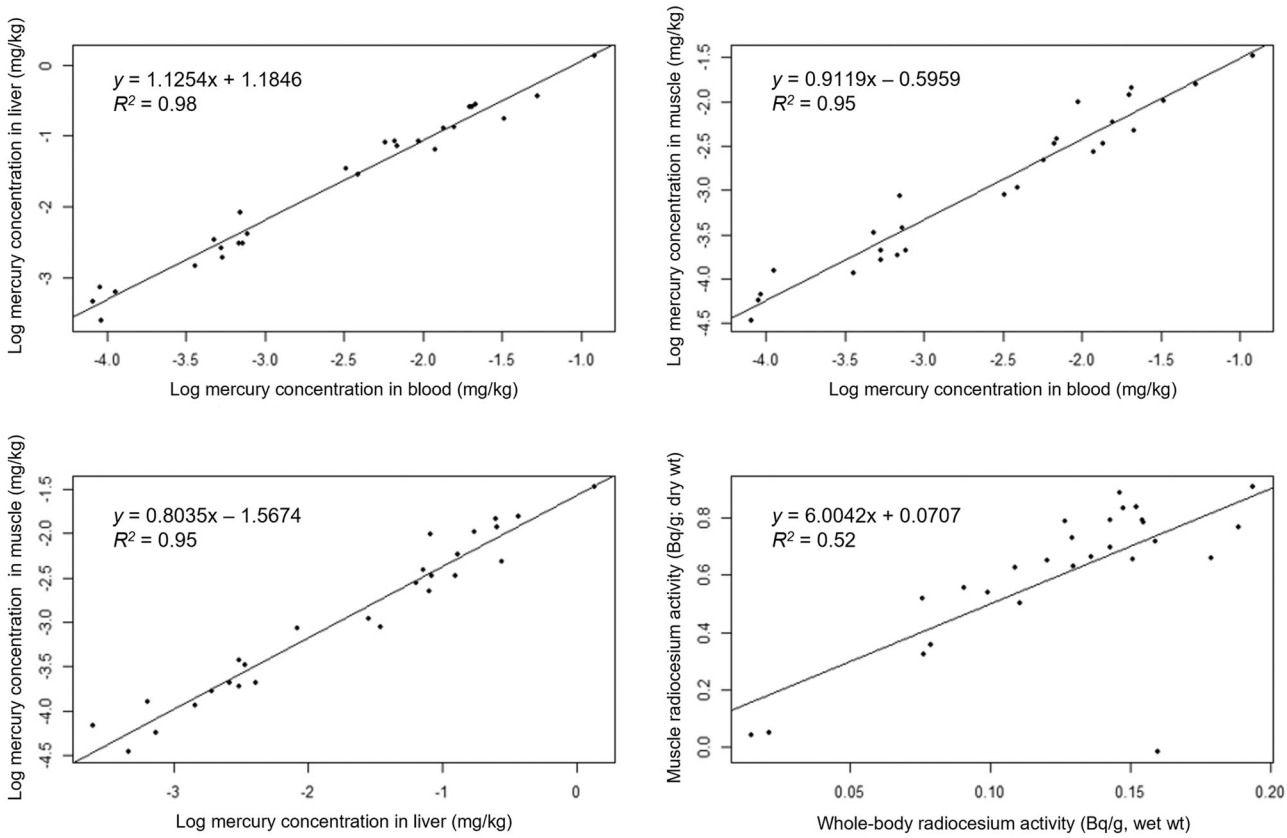

FIGURE 3: Linear relationships between total mercury concentrations and radiocesium activity between different tissue types from mallard ducks (*Anas platyrhynchos*) exposed to contaminants for known exposure periods in a contaminated wetland on the US Department of Energy's Savannah River Site in Aiken, South Carolina.

Fourmile Beaver Pond system likely contributed to observed differences in the accumulation of contaminants between species at our study site. Prior to collection of released birds, our mallards were often observed foraging below the beaver dam in shallow waters. With anthropogenic Hg present, combined with the constant desiccation and rehydration of wetland media in this riparian floodplain area, it is possible that higher methylation rates of Hg occurred in shallow areas subject to dry-out because these conditions were ideal for microbial sulfate reduction (Dmytriw et al., 1995; Feng et al., 2014). Ring-necked ducks, however, were always observed utilizing the deeper portion of the beaver pond, upstream of the dam, including areas with known contamination. While Hg and ^{137}Cs are present in portions of the areas where ring-necked ducks were observed (Kvartek et al., 1994; Xu et al., 2019), our data suggest exposure to Hg and ^{137}Cs were similar to background levels, and any ^{137}Cs was likely quickly excreted from the body given the lack of consistent exposure, similar to rapid ^{137}Cs attenuation observed in wood ducks (Fendley et al., 1977). For Hg, ring-necked ducks fed differently compared to mallards and this may have caused them to utilize different microenvironments within the study system that had possible differences in Hg methylation rates.

Likely foraging more heavily in contaminated areas compared to ring-necked ducks, mallards consistently accumulated both Hg and ^{137}Cs over the duration of our study. However, the levels of Hg found in blood never exceeded 0.2 mg/kg (wet wt, calculated from dry to wet ratios in Newman et al., 2011), placing them below background levels in a low-risk category for adverse health effects (Ackerman et al., 2016). Similarly, whole-body ^{137}Cs activity in our mallards from Fourmile did not exceed whole-body equivalent standards set by the EEC for human consumption (0.324 Bq/g, fresh mass; EEC, 1986) nor did their muscle tissues (0.600 Bq/g, fresh mass) using calculations for tissue moisture conversions from Kennamer et al. (2017). While these Hg concentrations and ^{137}Cs activity found in Fourmile mallards are considered lower risk at the time of our study, there may be seasonal variation in bioavailability not reflected in the present study. Furthermore, Hg accumulation in waterfowl tissues likely would have continued to increase beyond this 94-day exposure study, although the rate of accumulation was decreasing and appears to be approaching an asymptote toward the end of our study.

In the environment, MeHg fluctuates seasonally and can be affected by abiotic and biotic factors such as seasonal Hg deposition (Selin & Jacob, 2008), methylation associated with water and sediment chemistry and Hg speciation (Edmonds et al., 2012; Kim et al., 2015; Xu et al., 2019), microbial activity, and vegetative growth (Windham-Myers et al., 2014). Environmental components, such as acidity, dissolved organic matter in the water column, resuspension of sediments and

organic matter within the sediments, and decreased dissolved oxygen, all contribute to the increased methylation potential of Hg by sulfate- and/or iron-reducing bacteria (Edmonds et al., 2012; Kim et al., 2015; Xu et al., 2019). Once methylated, plant uptake through roots and leaf tissues during the growing season transports MeHg from abiotic into biotic components (Windham-Myers et al., 2014), and benthic organisms (e.g., crayfish, mussels, catfish) can uptake sediment particles or debris which can then be transferred into food webs for organisms consuming plant matter and benthic feeders. Furthermore, the seasonal availability of lower-trophic aquatic organisms, such as invertebrates, that are often consumed by waterfowl creates an additional route for MeHg biomagnification through aquatic and terrestrial food webs (Walters et al., 2020; Xu et al., 2019; Zhang et al., 2012). For mallards, dietary shifts associated with seasonal food availability (Hitchcock et al., 2021) could influence Hg exposure, resulting in seasonal variation in bioaccumulation. Notably, studies have reported that mallard diet shifts to 100% invertebrate prey during breeding (Swanson et al., 1985) before returning to a 69%–81% plant-based diet during the nonbreeding period (Sugden & Driver, 1980). This dietary shift could increase Hg exposure and accumulation, as seen in other avian species (Morrissey et al., 2010). As time progressed, our mallards transitioned into their breeding season, and Hg bioaccumulation may have been influenced as a result. While molting and egg laying are also important Hg excretion pathways in birds (Chételat et al., 2020), this is not applicable for mallards in our study considering we only used males during periods outside of the molting season.

Our findings further support the use of blood as a technique for estimating Hg contamination, allowing for the estimation of contaminant burdens in tissues. Similar to other studies (Eagles-Smith et al., 2008; Mallory et al., 2018; Sullivan & Kopec, 2018), we found Hg concentrations in whole blood had a strong, positive, linear relationship with both muscle and liver concentrations in the same bird. Furthermore, we found a similar relationship between whole-body and muscle activity of ^{137}Cs, where birds can be whole-body live counted to estimate ^{137}Cs activity within muscle tissues. These findings suggest nondestructive sampling techniques can be effectively used to estimate contaminant burdens within tissue types, with euthanasia of birds unnecessary. Such techniques could be employed to examine potential risks imposed on the health of wildlife or as a risk assessment toward the consumption of game species such as waterfowl.

Acknowledgment—The authors thank M.E. Winzeler, M.T. Hamilton, and M. Oberkircher for their help trapping and handling waterfowl. We also thank A.H. Lindell and R.A. Kennamer for their expertise and assistance with mercury and radiocesium analyses, respectively. Funding for the present study was provided by the US Department of Energy's Savannah River Nuclear Solutions – Area Completion Projects under Award No. DE-EM0004391 to the University of Georgia Research Foundation, and the National Science Foundation REU Program (Award No. 1460940).

Author Contributions Statement—All authors contributed equally to the participation of this manuscript in its entirety.

Data Availability Statement—All data pertaining to the present study is archived with the University of Georgia Savannah River Ecology Laboratory and may be made available from the corresponding author. Data are available on request due to the collection of sensitive contaminant data on a restricted US Department of Energy site.

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