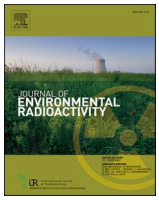
Journal of Environmental Radioactivity 262 (2023) 107164

Contents lists available at ScienceDirect

Journal of Environmental Radioactivity

journal homepage: www.elsevier.com/locate/jenvrad



Allometric-kinetic model predictions of radionuclide dynamics across turtle taxa

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ARTICLE INFO

Keywords:
Radionuclide
Contamination
Uranium
Plutonium
Turtle
Chelonian

ABSTRACT

Chelonians (turtles, tortoises, and sea turtles; hereafter, turtles) inhabit a wide variety of ecosystems that are currently, or have the potential in the future to become, radioactively contaminated. Because they are long-lived, turtles may uniquely accumulate significant amounts of the radionuclides, especially those with long half-lives and are less environmentally mobile. Further, turtle shells are covered by scutes made of keratin. For many turtle taxa, each year, keratin grows sequentially creating annual growth rings or layers. Theoretically, analysis of these scute layers for radionuclides could provide a history of the radioactivity levels in the environment, yet there are few previously published studies focused on the dynamics of radionuclide intake in turtles. Using established biochemical and ecological principles, we developed an allometric-kinetic model to establish relationships between the radionuclide concentrations in turtles and the environment they inhabit. Specifically, we calculated Concentration Ratios (*CRs* – ratio of radionuclide concentration in the turtle divided by the concentration in the soil, sediment, or water) for long-lived radionuclides of uranium and plutonium for freshwater turtles, tortoises, and sea turtles. These *CRs* allowed prediction of environmental concentrations based on measured concentrations within turtles or vice-versa. We validated model-calculated *CR* values through comparison with published *CR* values for representative organisms, and the uncertainty in each of the model parameters was propagated through the *CR* calculation using Monte Carlo techniques. Results show an accuracy within a factor of three for most *CR* comparisons though the difference for plutonium was larger with a *CR* ratio of about 200 times for sea turtles, driven largely by the uncertainty of the solubility of plutonium in sea water.

1. Introduction

Long-lived animals inhabiting sites contaminated with radionuclides can assimilate significant quantities into tissues (Whicker and Schultz, 1982). The amount of radionuclide accumulated into animals depends on numerous factors including the 1) concentrations in environmental media such as soil, sediment, and water, 2) bioavailability and transport rates of the radionuclide within each exposure pathway, 3) food intake rates, 4) exposure times and longevity of the animal, and 5) biokinetics of the radionuclides within the animals.

Radiobiological data, combined with exposure pathway models, helps predict the fate and transport of radionuclides through the environment and into animals. Commonly, analyses focus on lumped parameters such as the Concentration Ratio (*CR*), the ratio of radionuclide concentration in an animal to the concentration in the soil, sediment, or water, to estimate concentrations in animals or plants based on concentrations measured in the environment. The International Commission on Radiation Protection (ICRP) and others have summarized *CRs* for a set of 12 "representative organisms" (ROs), and these *CR* values enable radiation protection for non-human biota based on measurements of radionuclide concentrations in soil, sediment, and water (ICRP, 2008; ICRP, 2009; Beresford et al., 2008; Higley, 2017). Similarly, the Department of Energy (DOE) in the United States, which regulates radiation protection of the environment at DOE sites (DOE, 2020), uses broad categories of terrestrial animals and plants, riparian animals, and aquatic animals for their regulations. The DOE has published *CRs* and other transport parameter values for each of these categories (DOE, 2019).

The focus of this study is on radionuclide transport and intake for Chelonians (turtles, tortoises, and sea turtles; hereafter, turtles). Given

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https://doi.org/10.1016/j.jenvrad.2023.107164
Received 18 January 2023; Received in revised form 4 March 2023; Accepted 16 March 2023
Available online 28 March 2023

the paucity of radioecological data for turtles, if regulatory or investigative conditions required analysis of these taxa, current analyses would only focus on the closest category for each turtle species and its associated *CR* used to calculate a radiation dose from environmental measurements. For example, the closest regulatory categories for turtles found in DOE (2019) and in ICRP (2009) could be 1) the DOE's generic "riparian animal" or ICRP's "frog" as the RO for freshwater turtles, 2) the DOE category "terrestrial animal" or ICRP's "deer or a rat" as a RO for tortoises, and 3) an "aquatic animal" for the DOE category or ICRP's RO "flat fish" for sea turtles. While the DOE's and ICRP's radiation protection scheme is protective of many species (Charrasse et al., 2019), because of their uniqueness (e.g., ectothermic reptiles), the generic DOE categories and ROs may not accurately represent the diversity of bioaccumulation within turtle taxa. Though there have been several studies on uptake and retention of radionuclides in turtles, they are limited to a few species and radionuclides (Holcomb et al., 1971; Balazs et al., 1990; Hinton et al., 1992; Wood et al., 2010; Johansen et al., 2020). Therefore, there is not enough data on uptake and retention of radionuclides in a contaminated environment to specifically quantify accumulation in turtles and analyses require a taxon-specific approach for the pathway model to calculate radionuclide intake rates and assimilation across Chelonians.

1.1. Why turtles?

Studying radionuclide concentrations in turtles is particularly useful as an indicator of the concentrations in the local environment for several reasons. First, turtles broadly inhabit a variety of freshwater, desert, and marine ecosystems, and specifically, they are found within areas contaminated with radionuclides including environments surrounding nuclear power plants and sites associated with nuclear weapons production and testing (Towns, 1987; Balazs et al., 1990; Hinton et al., 1992; Johansen et al., 2020). Second, turtles are relatively long-lived, and in many cases, can live decades (Gibbons and Semlitsch, 1982). Longer residence times in contaminated areas results in continued accumulation of radionuclides and increasing body burdens. Longevity and limited movement of turtles (Gibbons, 1970, 1986) is especially important for continued accumulation of environmentally less-mobile radionuclides such as uranium and plutonium. Third, many long-lived radionuclides accumulate in bone, and the keratin shell overlaying this bony tissue will likewise assimilate radioisotopes (Whicker and Shultz, 1982). Importantly, shells of many turtles contain scute layers that represent annual growth rings. Thus, it may be possible to obtain histories of their radionuclide exposures through measurement of individual scute layers (Van Houtan et al., 2016). These factors make turtles ideal candidates for modeling radionuclide accumulation and retention rates.

1.2. Allometric relationships

Allometry, or biological scaling, provides the ability to describe quantitative relationships between metabolic processes in living organisms and their body size (Brown et al., 2004). Metabolic rates derive from the fundamental chemical processes and energy expenditure in animals required to meet basic functions to live, grow and reproduce. Because rates of food intake, respiration, and elimination are governed by metabolic rates, radionuclide intake rates and biological residence times are proportional to the mass of the animal (Higley et al., 2003a; Ulanovsky, 2016).

The biology, habitat, and behavioral characteristics among animal species contribute to their specific radionuclide dynamics, and it would be impossible to collect species-specific data for every possible organism. ICRP 136 (ICRP, 2017) suggests that one reasonable approach is to utilize established allometric relationships between body mass and metabolic processes to predict radionuclide intake and elimination rates. Though there are many factors that modify metabolic rates across the wide range of animal species, life stages, and specific environmental conditions (Hulbert, 2014), research indicates that these allometric relationships are reasonably accurate because they fundamentally rely on predictable biochemical reaction rates common to all living organisms (Brown et al., 2004). Additionally, it is important to note that these allometric relationships were derived using interspecies data, and here we assume they apply to intraspecies equally (Kleiber 1947).

Numerous characteristics of organisms, such as basal metabolic rates, rate of food intake, respiration rates, life spans, and home ranges, vary in proportion to the organism's mass (*M*). Each of these allometric relationships are generally described by a power function as shown in Eqn. (1) (Kleiber, 1947; Calder, 1984: Schmidt-Nielsen, 1984; West et al., 1997; Brown et al., 2004; West and Brown, 2005).

$$Y = Y_o M^b \quad (1)$$

where.

Y is the variable of interest,
Y_o is the normalization constant, and
b is the allometric exponent.

Specific for use in radionuclide intake in non-human biota, modification of Eqn. (1) facilitates calculation of rates of intake through ingestion (food, water, and soil) and inhalation, as well as prediction of elimination rates (Whicker and Schultz, 1982; ICRP, 2017; DOE, 2019).

A critical advantage of the allometric approach for the broad diversity of turtle taxa is that it allows predictions of radionuclide concentrations in turtles that vary in masses spanning, at least, three orders-of-magnitude and living in diverse environments such as deserts, riparian, and even marine ecosystems. Also, turtles consume a variety of foods including plants, insects, fish, jelly fish, and more, that make predictions for radionuclide intake rates complicated using traditional pathway analysis. Finally, being ectotherms, the basal metabolism rates for turtles change with temperatures, which drive changes in seasonal consumption rates and food sources.

1.3. Goal of this study

The goal of this study is to develop a radionuclide-specific pathway model for predicting radionuclide uptake and retention in turtles that span the spectrum of taxonomic diversity. To do this, we used an allometric-kinetic approach to estimate radionuclide concentrations of long-lived radionuclides in turtles and their shells. To capture the diversity of turtles and yet allow for comparisons, we categorized Chelonians as: 1) freshwater riparian turtles, 2) terrestrial tortoises, and 3) sea turtles. The range of masses within each of these categories was restricted to adults, and using these masses, the allometric-kinetic model provided estimates of the *CR* (i.e., ratio of turtle tissue concentration to concentration in the soil or sediment). We propagated the uncertainty in model parameters in the turtle pathway model to provide overall uncertainties in *CR* values. A sensitivity analysis determined critical parameters influencing *CR* values and provided information on key attributes in organisms contributing to higher radionuclide concentrations. For this study, we focused on uranium-238 and plutonium-239, two long-lived radionuclides commonly found at sites associated with nuclear power or nuclear weapons production and testing. Finally, we compared our model predictions in each category of turtle to known, published values.

2. Methods

2.1. Intake pathways and rates of elimination

Radionuclide retention is a function of rates of intake and elimination (Fig. 1) and is describable using first-order kinetic equations

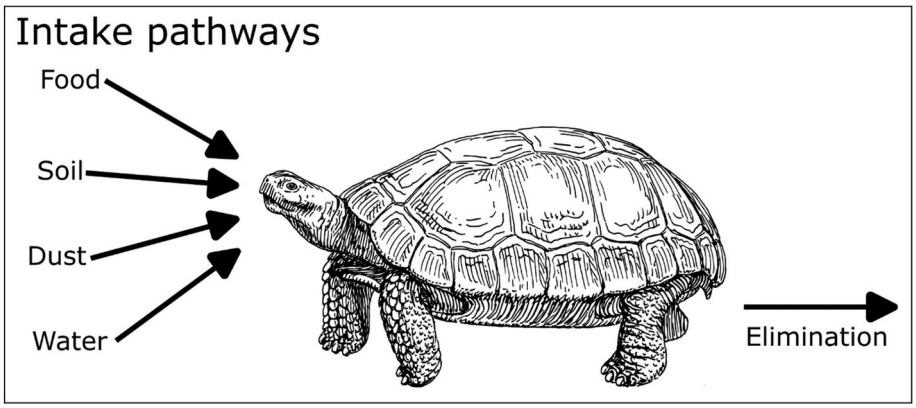

Fig. 1. Illustration of intake and elimination pathways for a turtle. Using allometric relationships, we use these pathways to mathematically predict the retention of radionuclides in turtles. Modified open-source tortoise line vector drawing from freesvg.org.

(Whicker and Schultz, 1982). Intake pathways include inhalation and ingestion of food, soil, and water. The elimination rate for each radionuclide is described by the sum of its biological and physical half-life. Because there are limited data on rates of intake (inhalation and ingestion) and elimination for turtles in the wild, we require allometric equations to predict these as a function of the mass of the turtle. Fundamentally, allometric equations derive from the power function in Eqn. (1). The framework, equations, and references for the following allometric equations are provided in Whicker and Shultz (1982), Higley et al. (2003a), and DOE Standard 1153 (DOE, 2019). Below are the equations used to predict the intake and elimination rates illustrated in Fig. 1.

2.2. Ingestion rate (R_{food})

The rate of food intake, R_{food} in units of g d^{-1}, as a function of mass (M_{kg}) can be predicted using Eqn. (2).

$$R_{food} = \frac{a}{dc}\left(70M_{kg}^{b}\right) \tag{2}$$

Here.

a is the ratio of active to maintenance metabolic rate,
b is the exponent in allometric relationship describing consumption as a function of body mass,
c is the caloric value of food intake [kcal/g],
d is the fraction of energy ingested that is assimilated, and
70 is an empirically derived value with appropriate unit conversions (Kleiber, 1947).

2.3. Soil ingestion rate (R_{soil})

The soil ingestion rate (g d^{-1}) is described as a fraction (f) of the food ingestion rate (R_{food}), as show in Eqn. (3).

$$R_{soil} = \frac{fa}{dc}\left(70M_{kg}^{b}\right) \tag{3}$$

The fraction of ingested soil mass by turtles was derived from the EPA Wildlife Handbook (EPA, 1993) and assumed to be 0.05, on a dry mass basis, of the food mass intake rate. We completed a conversion of wet to dry mass for the soil by assuming that the wet mass was, on average, twice that of the dry mass, so f was set at 0.1. Because turtles occupy habitats that include both soil and sediment, we assume that the wet mass fraction of 0.1 applied to both.

2.4. Water intake rate

The predicted rate of water intake (I_{water}) as a function of mass, M, is described in Eqn. (4) (Higley et al., 2003a).

$$I_{water} = 0.099M_{kg}^{0.9} \tag{4}$$

2.5. Respiration rate

Eqn. (5) provides the allometric equation for the respiration rate (R_b in units of m^3 d^{-1}) of turtles as a function of mass (kg) (Higley et al., 2003a)

$$R_b = 0.481M_{kg}^{0.76} \tag{5}$$

The respiration rate (m^3 d^{-1}) multiplied by the atmospheric dust loading (g m^{-3}) provides the mass intake rate of the turtle in g d^{-1} (Eqn. (6)). A reasonably conservative dust loading of 100 μg m^{-3} was applied as the mass concentration in air ($C_{mass\text{-}air}$) near the soil surface, but like other model parameters, was varied during uncertainty propagation.

$$R_{inhal} = R_b C_{mass-air} \tag{6}$$

2.6. Sum of intakes by mass and normalized radionuclide content

Mass intake rates (g d^{-1}) were summed (Eqn. (7)) and then converted to normalized radionuclide intake rates (Bq d^{-1}) assuming a soil mass concentration of 1 Bq g^{-1} and a water volume concentration of 1 Bq l^{-1} (Eqn. (8)). We applied specific Concentration Ratios (i.e., ratio of food/soil concentrations-$CR_{food/soil}$) to the rate of food intake (R_{food}) to calculate the radionuclide intake rate from the mass intake rate (Eqn. (8)). The radionuclide-specific values for $CR_{food/soil}$ were taken from DOE (2019) and IAEA (2010).

$$I_{total} = R_{food} + R_{soil} + R_{inhal} + I_{water} \tag{7}$$

$$R_{rad} = C_{soil}\left(CR_{food/\ soil}R_{food} + R_{soil} + R_{inhal}\right) + \left(I_{water} \times C_{water}\right) \tag{8}$$

2.7. Biological half-life (d)

For elimination rates, the biological half-life ($t_{1/2\text{-bio}}$) describes the time for radionuclide concentration in a turtle to decrease by 50%. The $t_{1/2\text{-bio}}$ is also related to the mass of the turtle, as shown in Eqn. (9).

$$t_{½-bio} = \alpha M_g^{\beta} \tag{9}$$

Where α and β are radionuclide specific and turtle mass, M_g, is in units of grams. For this work, α was set at 0.8 for both uranium and plutonium and β was set at 0.28 and 0.81 for uranium and plutonium, respectively (DOE, 2019). Beresford and Wood (2014) provide alternative approaches and values for the parameters in Eqn. (9) for reptiles and a limited set of radionuclides, but for uranium and plutonium in turtles

specifically, we used the default values in DOE Standard 1153 (DOE, 2019).

2.8. Uptake and retention of radionuclides through time

The general first-order equation for radionuclide uptake and retention of radionuclides (A_{rad}) is calculated using Eqn. 10

$$A_{rad}(t) = \frac{R_{rad}}{\lambda_{bio} + \lambda_{Phy}}\left(1 - e^{-\left(\lambda_{bio}+\lambda_{Phy}\right)t}\right) \quad (10)$$

where $A_{rad}(t)$ is the radioactivity assimilated into tissue through time, R_{rad} is the rate of radionuclide intake, λ_{bio} and λ_{phy} are the biological and physical decay loss rate constants, respectively, which are related to the biological ($t_{1/2\ bio}$) and physical ($t_{1/2\text{-}phy}$) half-lives (Eqn. (11)).

Here:

$$\lambda_{bio} = \frac{ln\ 2}{t_{\frac{1}{2}-bio}}, and \lambda_{phy} = \frac{ln\ 2}{t_{\frac{1}{2}-phy}} \quad (11)$$

Equation (10) is simplified by noting that the physical decay half-lives of plutonium and uranium are much longer than the biological half-lives and that the average lifetimes of the turtles (t) are generally longer than the biological half-lives. Therefore, we can calculate the equilibrium concentration in the turtle where t is long (compared to the biological half-life) as:

$$C_{equil}\left(Bq\ g^{-1}\right) = \frac{f_1 R_{rad}\left(t_{\frac{1}{2}-bio}\right)}{M_g\ ln\ 2} \quad (12)$$

Here, f_1 is the fraction of the radionuclide assimilated into the turtle's tissues. Default f_1 values were taken from DOE Standard 1153 (DOE, 2019). The selected f_1 values were 5E-02 and 1E-03 for uranium and plutonium, respectively. An additional partitioning of radionuclide content in the turtle shell can be made by multiplying C_{equil} by 0.15 and 0.5 for uranium and plutonium, respectively (ICRP, 2015; ICRP 2018). We expect the biokinetics in turtles are more complex than that described Eqn. (12), but without alternative data, we use it as a simplified but reasonable description of radionuclide assimilation in turtles. Substituting each intake pathways into Eqn. (12), the equilibrium concentration in an animal is described in Eqn. (13).

$$C_{turtle\ equil} = \frac{f_1}{M_g} * \left[C_{soil-sed}\left(\left(\frac{a}{dc}\left(70 * \frac{M_g}{1000}^{b}\right) * \left(CR_{p-s} + f\right)\right) + \left(x * \left(0.481 * \frac{M_g}{1000}^{0.76}\right)\right)\right) + \left(0.099\left(\frac{M_g}{1000}\right)^{0.9} * C_{water}\right)\right]\left[\frac{\alpha M_g^{\beta}}{\ln(2)}\right] \quad (13)$$

Measured concentration ratios are found in the literature for ROs and other species (Beresford et al., 2008; ICRP, 2009; IAEA, 2014; DOE, 2019). These CR values are generally calculated as the ratio of the radionuclide concentration in the organism (i.e., Eqn. (13)) to the concentration of either the soil/sediment, or in water (Eqn. (14)).

$$CR = \frac{C_{turtle-equil}}{C_{soil}-sed} or \frac{C_{turtle-equil}}{C_{water}} \quad (14)$$

Once obtained, CR values are used to predict concentration in turtles from measured environmental concentrations or, in reverse, to predict environmental concentrations from measured concentrations in turtles and/or their shells. It is important to note that Eqn. (14) predicts measured whole-body concentrations in turtles. An additional distribution factor can be used to partition the radionuclides to the shell. As described later in this paper, the fractions of radionuclides in the shells were 0.15 and 0.5 for uranium and plutonium, respectively (ICRP, 2015; ICRP, 2018).

2.9. Selection of radionuclides

Setting the concentrations of soil-sediment ($C_{soil\text{-}sed}$) and water (C_{water}) in Eqn. (13)–(1) Bq g^{-1} and 1 Bq L^{-1}, respectively, allowed CRs to be calculated for specific radionuclides. In this analysis, we are interested in radionuclides that 1) are long-lived relative to the life span of turtles (e.g., physical half-lives of decades or longer), 2) are found in locations that turtles inhabit with high soil-sediment concentrations, and 3) deposit and are retained in turtle shells and bones. For this study, we focused on uranium-238 and plutonium-239.

2.10. Selection of turtle categories

Chelonian taxa are diverse, and they inhabit broad ecosystem types ranging from freshwater ponds to dry, hot deserts to oceans. Thus, each taxon is expected to have specific behaviors and biology that could impact radionuclide intake rates that would be difficult to capture for each turtle species. While most allometric models are species-independent, in this study, an attempt was made to coarsely categorize turtle taxa for comparisons to CR values in the literature. The selected categories that allowed comparisons were: 1) riparian for freshwater turtles, 2) terrestrial for tortoises, and 3) marine for sea turtles.

2.11. Differences in model parameters among turtle categories

There were several important differences between the turtle categories that effected radionuclide uptake and retention in the model. First, there are adult mass differences assumed in the model with freshwater turtles having smaller mass (0.1–0.5 kg) compared to tortoises (2–20 kg) and sea turtles (10–150 kg). Second, the intake pathways varied. Intake pathways for freshwater turtles and tortoises included ingestion of food, soil and water plus inhalation of contaminated dust. In contrast, the sea turtle's only intake was through ingested food and sediment.

Finally, plant to soil concentration ratios differed between the turtle categories. The $CR_{plant\text{-}soil}$ values for freshwater turtles and tortoises were taken directly from DOE Standard 1153 (DOE 2019) and were 4E-3 for uranium and 1E-2 for plutonium. In contrast, the calculated $CR_{plant\text{-}sediment}$ values for sea turtle category were 7.3E-3 and 2.4E-2 for uranium and plutonium, respectively. These $CR_{plant\text{-}sediment}$ were calculated from $CR_{plant\text{-}water}$ values taken from ICRP 114 (ICRP 2009) for brown seaweed (2.9 E1 L kg^{-1} for uranium and 2.4 E3 L kg^{-1} for plutonium), which were then divided by k_d values for each of the radionuclides. The k_d values of 4 E3 L kg^{-1} and 1 E5 L kg^{-1} were the default values for uranium and plutonium, respectively (DOE 2019; IAEA 2004).

2.12. Uncertainty propagation and sensitivity analysis

Many of the variables used in the allometric-kinetic model have associated uncertainty. Through our Monte-Carlo analysis, we propagated these uncertainties through the allometric-kinetic model, similar to that found in Higley et al. (2003b). For this, each of the variables were assigned a triangular distribution with central values, and upper- and lower-bound values.

Table 1 provides the assumed distribution details for each of the parameters in Eqn. (13) with references for the values used. The software Crystal Ball[1] probabilistically selected values from these distributions and calculated the CR (Eqn. (12)). This calculation was repeated 10,000 times to obtain a distribution of CR values. Crystal Ball also provides a sensitivity analysis that calculates and compares the percent

[1] Oracle Crystal Ball Installation and Licensing Guide, Release 11.1.2.4.900, E70746-05, Copyright © 1988, 2020, Oracle and/or its affiliates. All rights reserved.

Table 1
Model parameters and distribution values used in Monte Carlo analysis.

Variable	Description	Lower Bound	Upper Bound	Likeliest	Reference(s)
a	ratio of active to basal metabolic rate	0.5	3	2	DOE (2019)
d	fraction of energy ingested that is assimilated	0.3	0.9	0.65	DOE (2019)
c	caloric value of food (kcal g^{-1})	4	9	5	DOE (2019)
b	Empirically determined for turtles/tortoises (consumption as a function of body mass)	0.675	0.8	0.75	DOE (2019); Ultsch (2013)
f	fraction of diet that is soil	0.05	0.2	0.1	EPA (1993)
x	airborne dust load (g m^{-3})	0.00005	0.0005	0.0001	DOE (2019)
CR_p-s:U	Concentration Ratio plant to soil for uranium-freshwater turtles and tortoises	0.0004	0.04	0.004	DOE (2019); IAEA (2010)
	Concentration Ratio: plant to sediment for uranium- sea turtles	0.0007	0.07	0.007	DOE (2019); IAEA (20
CR_p-s:Pu	Concentration Ratio: plant to soil for plutonium	0.00001	0.001	0.0001	DOE (2019); IAEA (2010)
	Concentration Ratio: plant to sediment for plutonium- sea turtles	0.0024	0.24	0.024	
C_soil	Normalized soil concentration-Constant (Bq g^{-1})	NA	NA	1	NA
C_water	Normalized water concentration-Constant (Bq L^{-1})	NA	NA	1	NA
f_1_U	fraction of intake assimilated into turtle for uranium	NA	NA	0.05	DOE (2019)
f_1_{Pu}	fraction of intake assimilated into turtle for plutonium	NA	NA	0.001	DOE (2019)
$F_{shell\text{-}U}$	fraction assimilated into turtle shell- uranium	NA	NA	0.15	ICRP 137 (ICRP 2015)
$F_{shell\text{-}Pu}$	fraction assimilated into turtle shell- plutonium	NA	NA	0.5	ICRP 141 (ICRP 2018)
Alpha (α)	scaling constant related to bio elimination	NA	NA	0.8	DOE (2019)
$beta_U$ (β_U)	scaling exponent related to bioelimination-uranium	NA	NA	0.28	DOE (2019)
$beta_{Pu}$ (β_{Pu})	scaling exponent related to bioelimination-plutonium	NA	NA	0.81	DOE (2019)
M_g–*FW turtle*	adult body weight in g	100	1000	500	Ultsch (2013)
M_g-*tortoise*	adult body weight in g	2000	200000	20000	Ultsch (2013)
M_g- *Sea turtle*	adult body weight in g	10000	400000	150000	Ultsch (2013)

of the variability in the *CR* distribution that is explained by each of the individual variables. This allows ranking of the variables in order of importance to the prediction of *CR* values.

2.13. Validation of models

We compared published *CRs* from DOE Standard 1153 (DOE 2019) and ICRP 114 (ICRP 2009) to calculated *CRs* from the allometric model for both uranium and plutonium in freshwater turtles, tortoises, and sea turtles. Additionally, we used Pu-239 concentrations in the shells of green sea turtles and algae collected from Johnson Atoll (Balazs et al., 1990) to calculate a measured $CR_{shell/algae}$ value. The allometric-kinetic model was used to calculate a similar $CR_{shell/algae}$ for comparison.

2.14. Temperature dependence of allometric equations

Because biochemical reaction rates are temperature dependent, the allometric equation (Eqn. (1)) should also contain a temperature correction where appropriate (Brown et al., 2004). While warm-blooded endotherms maintain a relatively constant body temperature, the internal body temperature for ectotherms is more likely influenced by swings in outdoor temperatures. The correction for temperature, T_c, is:

$$T_c = e^{-\frac{E}{kT}} \quad (15)$$

where.

E is the activation energy (assumed to be 0.65 eV),
k is Boltzmann's Constant (8.617E-5 eV K^{-1}), and
T is absolute temperature in degrees Kelvin.

Although turtles are ectotherms, they adjust behavior in ways to help regulate their body temperature for steadier metabolic rates. For example, in colder temperatures, freshwater turtles bask thus raising their body temperature significantly, or during hot summer days, they stay in cooler waters. Similarly, tortoises seek shade in hot weather and hibernacula during colder periods, and sea turtles limit the boundaries of their migratory range and depth in the ocean column to seek habitable temperatures. Combined, these behaviors moderate the range of possible body temperatures. Studies show that body temperature of turtles in each of the categories in this study ranged from 10 °C to 30 °C with an annual mean of roughly 20 °C, and this temperature distribution was relatively consistent among each turtle category (Grayson and Dorcas, 2004; Nussear et al., 2007; Patel et al., 2021). To evaluate the relative magnitude of the temperature corrections, we used the lowest, highest, and the annual mean body temperatures to calculate T_c (Eqn. (15)).

3. Results and discussion

3.1. Concentration ratios

The parameter distributions in Table 1 were used in Eqns. (13) and (14) then iterated 10,000 times using Monte Carlo techniques to generate distributions for the $CR_{turtle/soil\text{-}sed}$ for each category of turtle and the results are presented in Table 2. Table 2 provides the mean, standard deviation, and percentiles for $CR_{turtle/soil\text{-}sed}$ values for both uranium and plutonium.

3.2. Comparison of $CR_{turtle/soil\text{-}sed}$ values across turtles and radionuclides

These results show that for smaller turtles, i.e., the riparian/freshwater category, the $CR_{turtle/soil\text{-}sed}$ values for uranium are slightly higher than for plutonium, but this was not true for the larger tortoises and sea turtles (Fig. 2). Specifically, *CRs* proportionately scale with mass for plutonium but much less so with uranium. Predicted mean *CRs* for larger turtles are about a factor of 1.2 higher for uranium relative to the smallest taxon while *CRs* for plutonium increased by a factor of more than 40 with increasing mass.

This is explained by the fact that while the assimilation rate of uranium into turtle tissue is much higher than for plutonium (Fig. 3), the biological half-life for plutonium is longer than for uranium (Fig. 4), and the difference in biological half-life is especially pronounced for larger turtles. Given this, Eqn. (9) shows that for long-lived radionuclides the longer the biological half-life the more time is needed to reach equilibrium concentrations in the turtle. Fig. 5 provides an example of this relationship for a 10 kg turtle living in an environment with ubiquitous soil and water concentrations of 1 Bq g^{-1} and 1 Bq L^{-1}, respectively. In this case, equilibrium for uranium is achieved in about 100 days, but for plutonium it will take much longer (>10,000 days), and that after about 1000 days, the plutonium concentrations exceed that for uranium. These results demonstrate that because allometric relationships predict longer life spans for larger tortoises and sea turtles, there is more time for plutonium to concentrate in these Chelonians.

Table 2
Means, standard deviations, and percentile $CR_{turtle/soil\text{-}sed}$ values for uranium and plutonium within each category of turtle.

Parameter	Riparian/Freshwater Turtle		Terrestrial/Tortoise		Marine Turtle	
	Uranium	Plutonium	Uranium	Plutonium	Uranium	Plutonium
Mean	1.96E-03	9.65E-04	2.23E-03	1.46E-02	2.40E-03	4.24E-02
Std. Dev	9.34E-04	5.17E-04	1.07E-03	9.29E-03	1.22E-03	2.61E-02
P100	2.62E-04	1.05E-04	3.26E-04	7.28E-04	2.09E-04	3.45E-03
P90	9.29E-04	4.17E-04	1.07E-03	5.30E-03	1.10E-03	1.61E-02
P80	1.18E-03	5.38E-04	1.35E-03	7.09E-03	1.41E-03	2.16E-02
P70	1.39E-03	6.40E-04	1.58E-03	8.82E-03	1.66E-03	2.64E-02
P60	1.60E-03	7.45E-04	1.80E-03	1.05E-02	1.90E-03	3.12E-02
P50	1.81E-03	8.59E-04	2.04E-03	1.24E-02	2.16E-03	3.63E-02
P40	2.02E-03	9.82E-04	2.29E-03	1.45E-02	2.47E-03	4.23E-02
P30	2.29E-03	1.13E-03	2.60E-03	1.72E-02	2.81E-03	4.94E-02
P20	2.64E-03	1.33E-03	2.99E-03	2.08E-02	3.26E-03	5.93E-02
P10	3.21E-03	1.65E-03	3.65E-03	2.70E-02	3.96E-03	7.59E-02
P0	8.38E-03	4.22E-03	9.96E-03	7.95E-02	1.24E-02	2.43E-01

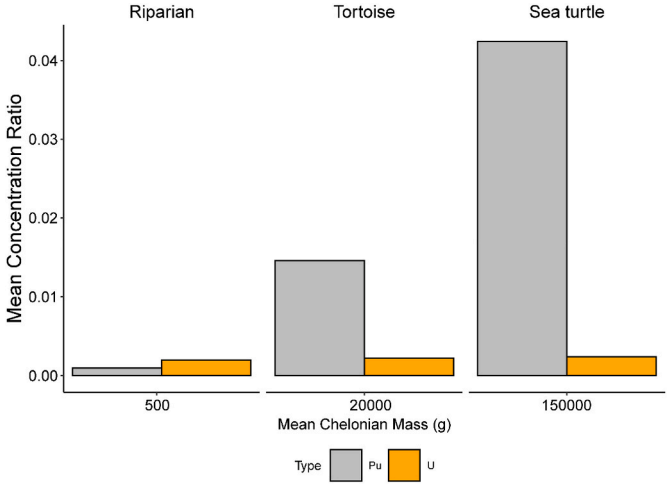

Fig. 2. Relationship of $CR_{turtle\text{-}soil/sed}$ with average mass of turtle for uranium and plutonium.

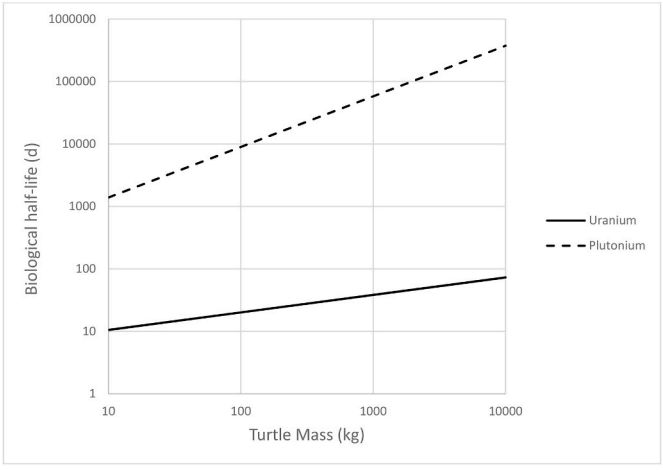

Fig. 4. Comparison of biological half-life for plutonium and uranium in turtles as a function of mass (see Eqn. (8)). Note that the biological half-life is much longer for plutonium than uranium and more strongly dependent on body mass.

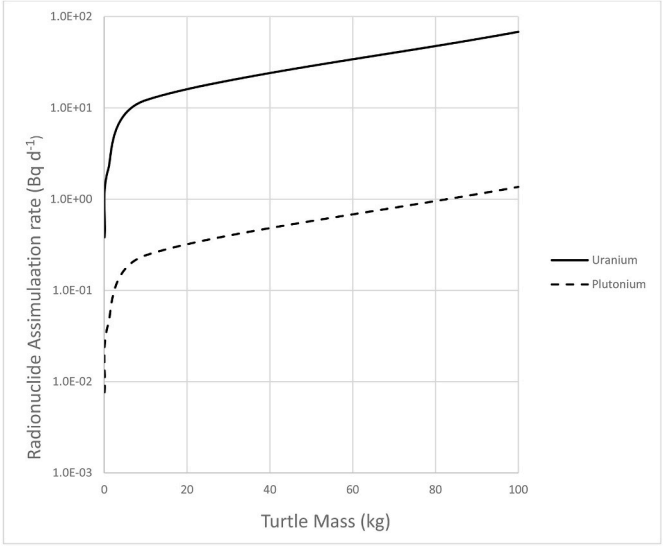

Fig. 3. Assimilation rate of radionuclides into tissue as a function of turtle mass based on allometric model for uranium and plutonium.

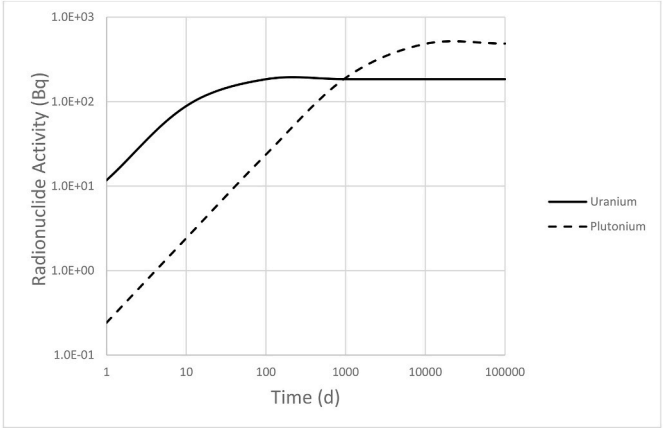

Fig. 5. Example of the predicted uranium and plutonium concentrations through time in the tissue of a 10 kg turtle living in an environment with soil and water concentrations of 1 Bq g^{-1} and 1 Bq L^{-1}, respectively.

3.3. Results from the sensitivity analysis

We provide a summary of the results of the sensitivity analysis across turtle taxa and radionuclides in Table 3. In all cases, the results of the Monte Carlo analysis show that the *CR* is relatively sensitive to changes

Table 3
Sensitivity analysis results- Percentage of variance explained by key variables. Positive values reflect proportional relationships while negative numbers show inverse relationships.

Parameter	Riparian Turtles		Tortoises		Sea Turtles	
	Uranium %	Plutonium %	Uranium %	Plutonium %	Uranium %	Plutonium %
Ratio of active to basal metabolic rate	39.1	31.6	37.1	20.3	36.8	25.1
Body mass (g)	<1	15.7	<1	42.5	<1	20.5
CR (food/soil-sed)	<2	<1	1.9	<1	5.5	20.4
Fraction of energy assimilated from food	−18	−15.8	−18	−9.4	−17.7	−12.9
Caloric value of food (kcal)	−13.6	−10.9	−13.1	−7	−12.3	−8.5
Fraction of diet that is soil	26.9	25.7	24.8	18.1	20.8	7.4
Food intake rate (g/d)	<1	<1	4.9	2.6	6.6	5.3

in 1) the ratio of the rate of active to basal metabolism (variable *a*), 2) fraction of diet that is soil (*f*), and 3) body mass (*M*), especially for plutonium. The amount of radionuclide in the turtles is dominated by the food and soil ingestion pathway, which is driven by body mass, whereas rates of radionuclide intake by inhalation and water ingestion are orders-of-magnitude lower than ingestion, which explains these results. The soil ingestion amount is particularly important since soil-bound radionuclides are directly ingested whereas only a fraction of the radionuclides in soil assimilate in food sources (e.g., plants, insects) that is later ingested. Mathematically, variables *a* (active to basal metabolism), *c* (caloric value of the food) and *d* (fraction of calories absorbed into the turtles) are all direct multipliers in Eqn. (13) with variable *a* being directly proportional and variables *d* and *c* being inversely proportional to the food intake rate. Of these variables, the range of values used in the Monte Carlo analysis varied from 0.5 to 3 (factor of 6) for variable *a*, 0.3 to 0.9 (factor of 3) for variable *c* and from 4 to 9 (factor of 2.5) for variable *d*. Combined with the multiplicative relationship with food intake, the larger range in values for variable *a* explains why the CR was sensitive to this variable for all turtles and radionuclides. This analysis suggests that more research is required to narrow the range of these variables and increase the predictive power of Eqn. (13). Finally, reptiles, such as turtles, in some cases have higher field metabolic scaling than other animal taxa (Nagy et al., 1999), and this should be considered in future work to refine model predictions.

The other interesting difference in the sensitivity analysis is the apparent lack of sensitivity of the *CR* to changes in body mass for uranium. While body mass explained 16–43% of the variation in the *CR* for plutonium, there was no similar relationship with uranium. Body mass was similarly important for intake rates of both uranium and plutonium; however, the mathematical relationship for tissue retention of the two radionuclides with body mass were very different, as described previously (see Figs. 4 and 5). In this case, the equilibrium concentrations of uranium in turtles (Eqn. (13)) would not be expected to vary strongly with increasing mass of the turtles.

3.4. Validation of the model

We tested the model results by comparing model-predicted *CR* with published *CR* values, though there are few published *CRs* for uranium and plutonium in turtle taxa. However, *CRs* have been published for generic/representative terrestrial, freshwater, and marine animals (IAEA, 2004; IAEA, 2014; ICRP, 2009; DOE, 2019), and these provide a comparison to the *CRs* calculated for the turtles in this study. The results of the comparisons are provided in Table 4. Note that we completed the *CR* comparison for plutonium in sea turtles using both a literature value for a "similar" RO and data from Green Turtles and collocated food sources of algae from Johnston Atoll (Balazs et al., 1990). The ratios of the model-predicted *CRs* to the published values were generally within a factor of three for all the uranium *CRs*, but there were greater differences between modeled and published *CR* values for plutonium especially for the tortoise and the sea turtle that were a factor of 36 and 200 times greater than the published values, respectively. The larger difference in the *CR* ratio for Pu in the sea turtle is likely the result of the k_d value used in the model to predict concentrations in water from the normalized soil concentration of 1 Bq g^{-1}. This additional calculation adds to the uncertainty because of the large range of possible k_d values. For example, in the sea turtles, the k_d value recommended for plutonium in ICRP 114 (ICRP, 2009) and DOE Standard 1153 (DOE, 2019) is 1E+05 L kg^{-1}, but the IAEA Technical Report 472 (IAEA, 2010) recommends a k_d value of 740 L kg^{-1}, which is in the range of k_d values (2 E+2 to 2E+7 L kg^{-1}) listed in DOE (2019). Using the lower k_d value results in a comparative ratio of the *CR* of 1.5 for the marine turtle. Table 4 provides both results.

As an additional validation, the model results were compared to the CRs(turtle to water) reported in Table 10 in Wood et al. (2010), which are 5.92E3 L kg^{-1} and 1.86E2 L kg^{-1} for plutonium and uranium, respectively. Because these CRs are based on water measurements, an additional conversion is needed to make them based on soil concentrations, thus allowing comparison to the model predictions. Assuming the default k_d values of 1E5 L kg^{-1} for plutonium and 4E3 L kg^{-1} for uranium, then the CR (turtle/soil) values from Wood et al. (2010) would be 5.92E-2 and 4.5 E−2 for plutonium and uranium, respectively. The

Table 4
Validation of Allometric-Kinetic model predictions. Comparison of modeled CRs to published CR values.

Turtle Category	Radionuclide	Published CR (mean)	Model CR	Ratio: Model to published	Source of Concentration Ratios
Riparian turtle[a]	Uranium	1.6E-03	1.9E-03	1.2	DOE 1153: Riparian Animal to Sediment
	Plutonium	3.6E-04	9.7E-04	2.7	DOE 1153: Riparian Animal to Sediment
Tortoise[a]	Uranium	1.7E-03	2.2E-03	1.3	DOE 1153: Terrestrial Animal to Soil
	Plutonium	4.1E-04	1.5E-02	35.6	DOE 1153: Terrestrial Animal to Soil
Sea turtle[a]	Uranium	4.0E-03	2.4E-03	0.6	ICRP 114: Ratio Flatfish/water (4 L kg^{-1}) Table 4.4 and k_d = 1E3 L kg^{-1} (IAEA 422 Table II)
	Plutonium	2.1E-04	4.2E-02	200.0 (1.5)[b]	ICRP 114: Ratio Flatfish/water (21 L kg^{-1}) Table 4.4 and k_d = 1E5 L kg^{-1} (IAEA 422 Table II).
		1.3E+00	1.7E+00	1.29	Shell/algae concentration ratios-Johnson Atoll data for Green Sea Turtle derived from Balazs et al. (1990)

[a] *CR* -ratios of concentration in turtle (Bq/g) to the concentration of the soil/sediment (Bq g^{-1})- unitless.
[b] Ratio based on k_d value of 740 for plutonium [IAEA Technical Report 472 (IAEA 2010)].

modeled values in Table 2 of this paper for freshwater turtles are lower than the Wood et al. (2010) values by factors of 0.02 for plutonium and 0.04 for uranium. As with the marine turtle comparison above, given the large ranges for possible k_d values and the limited data set, the relative uncertainty of this comparison is large.

3.5. Concentrations in turtle bony shell

Many long-lived radionuclides deposit in bone, and plutonium and uranium are examples. Therefore, we expect that these would accumulate in the bony shell (carapace/plastron) of the turtles with the amount deposited varying based on the biokinetics of the radionuclide. Of course, there are no biokinetic models for radionuclides in turtles as there are for humans, and often, the only tissue samples remaining from turtles collected decades earlier are the shells with or without the overlying keratin scute. Therefore, we used the biokinetic model data for humans (ICRP, 2015; ICRP, 2018) to estimate the concentration in the shell by multiplying the total turtle tissue concentration by the fractional amount deposited in bone. The fraction of these radionculides in bone relative to the amount other tissues used were 0.15 and 0.5 for uranium and plutonium, respectively. In this manner, concentrations in soil/sediment during the turtle's exposure can be calculated as the radionuclide concentrations in the turtle shell, divided by the ratio of the concentration of a radionculide in bone relative to the amount other tissues, then multiplied by the applicable *CR*.

3.6. Temperature dependence of allometric equations

To evaluate the relative magnitude of the temperature corrections throughout the seasonal range of body temperatures (Eqn. (15)), the lowest (10 °C), highest (30 °C), and annual mean (20 °C) body temperatures were used to calculate T_c (Grayson and Dorcas, 2004; Nussear et al., 2007; Patel et al., 2021). We calculated ratios of the relative corrections as the ratio of T_c values for the range of body temperatures. The calculated ratio of the annual mean T_c to the low temperature T_c was 2.5, the ratio of the upper/mean T_c was 2.3, and the ratio of the lower to upper T_c values was 5.8. Given that the goal for this study was to calculate *CRs* averaged over many seasons, the factor of 2 difference between the mean and the two extreme temperatures was not deemed significant enough to adjust the *CR* values.

4. Conclusions

We developed and tested a radionuclide-specific allometric-kinetic model for predicting radionuclide uptake and retention in turtles spanning a broad range of taxa. This model provided a calculation of *CR* values to predict radionuclide concentrations in three general types of turtles given uranium and plutonium contamination in the environment.

To evaluate the accuracy of the model, we compared *CRs* from the model to published *CR* values for representative animals. The comparisons showed reasonable agreement with some differences noted, especially for the *CRs* for plutonium since the k_d values in the literature vary by more than several orders-of-magnitude. A sensitivity analysis of the pathway model showed that the most sensitive parameters in Eqn. (13) were *a*-the ratio of the active to basal metabolic rate, *M*-the mass of the turtle and *f*-the fraction of the diet that is soil/sediment. Therefore, more specific information on the mass of the individual Chelonian under investigation would help narrow down the range of metabolic rates (Ultsch, 2013) in future studies.

The main conclusion of this work is the calculation of normalized *CRs* for a wide variety of turtles ranging from small-to large-sized taxa. These *CR* values thus provide either a concentration prediction in turtles based on environmental concentrations or, in the reverse, predictions on past environmental concentrations based on measurements of turtle tissue concentrations. We selected uranium and plutonium radionuclides in this study, but the method and approach are broadly applicable for other radionuclides and other organisms, if properly parameterized.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

No data was used for the research described in the article.

Acknowledgements

Research presented in this article was supported by the Laboratory Directed Research and Development program of Los Alamos National Laboratory under project number 20210220 ER. We thank our project team members for helping make this research possible. A special thanks to George Balazs for providing access to references.

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