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Rubidium and Cesium Kinetics and Tissue Distributions in Channel Catfish (*Ictalurus punctatus*)

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Abstract. We used a two-compartment, clearance volume-based model to examine rubidium and cesium pharmacokinetics in channel catfish (*Ictalurus punctatus*) after intravascular administration. We compared the apparent volumes of distribution in the central and peripheral compartments and the intercompartmental and whole-body clearances of both metals at 20.0 °C and 27.5 °C. Biological half-times of Rb were 15 to 16 d at both temperatures, but Cs biological half-times averaged 101 d and 85 d at 20.0 °C and 27.5 °C, respectively (5 to 7 times longer than those of Rb in the same individual). Both the intercompartmental and total body clearances of Rb were also 6 to 7 times greater than those of Cs. The apparent volumes of distribution for Rb in the central compartments were twice those of Cs and remained constant with temperature. The apparent volumes of distribution of both elements in peripheral compartments were large compared with their corresponding central compartments, and decreased by a similar extent with increased temperature. Cesium tissue to blood ratios were greatest for white muscle, with more than 85% of the Cs present in this tissue. Partitioning of Cs in peripheral tissues apparently decreased with increased temperature conditions. Our results indicate that application of pharmacokinetic modeling techniques can enhance studies of radionuclide kinetics by helping to identify rate-limiting processes within individuals that may control uptake and elimination.

Keywords: rubidium; cesium; kinetics; clearance-volume model; fish

Introduction

Environmental releases of radionuclides are an unfortunate reality of modern existence. The explosion and fire at the Chornobyl (Chernobyl) Nuclear Power Plant a decade ago produced significant radionuclide fallout in many areas of the world, including locations thousands of kilometers distant from the reactor (Medvedev, 1990; Kryshev, 1992). Although much of the initial radionuclide release consisted of short-lived fission products, large quantities of longer-lived and biologically active radionuclides such as ^{137}Cs (radioactive half-life, $T_p = 30.2$ y) persist in many of these areas. Because ^{137}Cs is readily accumulated and concentrated in skeletal muscle, consumption of contaminated fishes and other wildlife can be an important pathway for human exposure (Forseth et al., 1991). Consequently, there is renewed interest in the fate of radionuclide contaminants in aquatic ecosystems, and new studies of the importance of fishes as indicators of the presence of radionuclides and/or their potential contribu-

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tion to human dose commitments are emerging (e.g., Korhonen, 1990; Forseth *et al.*, 1991; Koulikov and Ryabov, 1992).

Some recent studies of aquatic vertebrates (e.g., Evans, 1988; Peters and Brisbin, 1996) have combined physiological and ecological models to predict ^{137}Cs kinetics. However, most radiocesium kinetics studies continue to emphasize direct measurements of biological half-time (T_b) in the field or laboratory. These studies have revealed a wide range of T_b values for ^{137}Cs in freshwater fishes (Table 1). The T_b's of ^{137}Cs of individual species are affected by factors that influence metabolic rate, decreasing with increasing temperature and decreasing mass (e.g., Kevern, 1966; Gallegos and Whicker, 1971; Fagerström, 1977; Mailhot *et al.*, 1989; Ugedal *et al.*, 1992). Because many elimination studies did not adequately describe the sizes of the animals used, or the water conditions under which measurements were made, it is difficult to discern patterns of Cs elimination across taxa. In a recent effort to produce a general model of Cs elimination in fishes, Rowan and Rasmussen (1995) reviewed published studies in an effort to produce a multiple regression model of radiocesium elimination as a function of temperature and body mass. Of the twenty studies that they reviewed, however, only five contained sufficient information on both temperatures and body masses to be included in their model (with most of the data drawn from a single study of brown trout). The authors also noted differences the T_b's of acutely- and chronically-contaminated fishes. As a consequence, the present inability to predict the radionuclide kinetics of fishes after contamination events perpetuates the necessity

Table 1. Whole-body elimination half-times (T_b) of alkali metals in freshwater fishes

Species	Wet mass (g)	Temperature (°C)	T_b (d)	Reference
K				
Lepomis macrochirus	50–70	17	37	Kolehmainen (1972)
Esox lucius	100–1500	8–10	168–248	Carlsson (1978)
Rb				
Carassius auratus	3–11	10–24	8.75–181	Peters (1996)
Ictalurus punctatus	122–278	20	16.4	This study
		27.5	15.8	This study
Cs				
Cyprinus carpio	137*	12.5	174	Kevern (1966)
		20	98	Kevern (1966)
Leuciscus rutilus	NA	10–20	55–100	Häsänen et al. (1967)
Perca fluviatilis	NA	10–20	175–200	Häsänen et al. (1967)
Salmo iridaeus	NA	10–20	25–80	Häsänen et al. (1967)
Onchorhynchus mykiss	30–200	5–18.3	34–92	Gallegos and Whicker (1971)
Lepomis macrochirus	0.5–120	15.8	86–187	Kolehmainen (1972)
Esox lucius	100–1500	8–10	402–599	Carlsson (1978)
Salmo trutta	NA	NA	124[b]	Forseth et al. (1991)
Salvalinus alpinus	NA	NA	150[b]	Forseth et al. (1991)
Hypophthalmichthys molitrix	500–7000	NA	44–231	Koulikov and Ryabov (1992)
Salmo trutta	12–456	4.4–15.6	104–564	Ugedal et al. (1992)
Micropterus salmoides	130–423	15	322	Peters and Newman (1999)
		20	225	Peters and Newman (1999)
		26	140	Peters and Newman (1999)
Ictalurus punctatus	122–188	20	154	This study
		27.5	84.6	This study

[a] Mean estimated indirectly from other parts of the study (Rowen and Rasmussen 1995)

kinetics and their concomitant health and ecological risks.

Rb and Cs are commonly considered to be biochemical analogs of K and to possess similar physiological behaviors (Whicker and Schultz, 1982). However, this appears to be true only for assimilation: Cs T_b tends to be greater than that of K in the same animal and both Cs concentrations and Cs/K ratios often increase with increasing trophic level by a factor of 2 to 3 (Reichle and Nelson, 1970; Whicker and Schultz, 1982). Whole-body Rb kinetics are commonly considered to be more similar to the kinetics of K, and Rb radionuclides are often employed as K tracers in metabolic studies (Dørup and Clausen, 1994). However, few studies are available that describe the elimination kinetics of Cs compared with other alkali metals or its pattern of distribution in fishes.

We believe that a better understanding of the physiology of alkali metals may improve the linkage between physiological and ecosystem models. Pharmacokinetic modeling techniques can aid in this effort by providing more detailed information on internal compartmentalization kinetics than can be obtained through whole-body elimination measurements alone. This information includes comparisons of the apparent volumes of distribution, defined as the volumes of the compartments (e.g., blood and well-vascularized tissues compared with peripheral tissues) in which the total amount of contaminant is associated. Such models can also estimate the clearances (i.e., the volumes cleared of contaminant per unit time) of the contaminant from, and exchanges between, these tissue compartments. This information can be combined with data on the distribution patterns of the contaminant within various tissues and organs to improve predictions of contaminant kinetics and concentrations in edible tissues.

We therefore measured the whole-body elimination of intravascularly administered Rb and Cs in a common food fish, the channel catfish, *Ictalurus punctatus*. We compared the kinetics of these two elements with a pharmacokinetic model fitted to combined whole-body and blood elimination data, which allowed us to estimate their intercompartmental clearances and apparent volumes of distribution. We also quantified the distribution of ^{137}Cs within various tissues and organ compartment.

Methods

Experimental animals

We obtained channel catfish (122–188 g) of both sexes from a local supplier (Orangeburg Aquaculture, Cordova, SC) and maintained them in 400-L fiberglass tanks (LS 700, Frigid Units, Toledo, OH). We used reconstituted hard water (United States Environmental Protection Agency 1978) prepared from the following reagent-grade salts (mg L^{-1}): $NaHCO_3$ (192), $CaSO_4 \cdot H_2O$ (120), $MgSO_4$ (120), and KCl (8), and to which an additional 0.1% (w/v) of NaCl was added. The median pH of this water was 7.8 (range 7.6–8.0, $n = 12$), and the total alkalinity and hardness were 110–120 and 160–180 mg L^{-1} as $CaCO_3$, respectively. Upon arrival, the fish were treated for 2 h in a 250 μg L^{-1} malachite green solution and then maintained at the experimental temperatures (20.0 or 27.5, $\pm$1.0 °C) for at least 4 wk before beginning the experiments. The fish were fed a commercial high-protein ration (Rangen soft moist pellets, Rangen, Inc., Buhl, ID), approximately 2% of their wet mass three times per week.

Radionuclide injections and blood sampling

We administered radiotracers (Amersham Corporation, Arlington Heights, IL) of Rb (^{86}Rb, T_p = 18.7 d) and Cs (^{137}Cs) to 14 fish through a dorsal aortic cannula. We used surgical procedures similar to those described by Kitzman *et al.* (1988) and Stehly and Plakas (1993), except that we used 150 mg L^{-1} methane tricaine sulfonate (MS-222) for the surgical anesthetic, an 18-gauge intravenous catheter (Angiocath, Becton Dickinson, Sandy, UT) to guide the cannula into the dorsal aorta, and 28-gauge Teflon tubing (Zeus Inc., Raritan, NJ) as the cannula material. The cannula was exteriorized through a hole in the rostrum. After surgery, each fish was immediately returned to its acclimation tank and placed in an individual compartment. The end of the cannula

was attached to a 1-mL syringe, which floated at the water surface. After 24 h, the cannulated fish were transferred to covered 19-L (45-cm diameter) polyethylene cages (Fig. 1), which were perforated to permit water exchange. The cages floated freely in covered 568-L polyethylene tanks (Rubbermaid Commercial Products, Inc., Winchester, VA). Each tank contained 6–7 cages, with less than 3 g fish L^{-1} water.

The fish were injected with a mixture of 3.486 kBq μL^{-1} of ^{86}Rb (specific activity, 81.5 kBq ^{86}Rb μg^{-1} Rb) and 1.440 kBq μL^{-1} of ^{137}Cs (specific activity, 11.8 kBq ^{137}Cs ng^{-1} Cs) dissolved in a modified Cortland physiological saline (Houston, 1990, with the albumin omitted). Each fish received an injection volume of 500 μL kg^{-1} of radionuclide solution, and the cannula was immediately rinsed three times with blood from the fish. These injections, after complete mixing in the blood, would elevate blood Rb^+ concentrations by about 8.4 nmol mL^{-1} (25.61 μg kg^{-1}) and Cs^+ concentrations by 12 pmol mL^{-1} (588 ng kg^{-1}). The added Rb^+ was therefore less than 0.27% of the molarity of K^+ expected to be present in the blood (3.14 μmol mL^{-1}; Cameron 1980), and the amount of added Cs^+ was 0.0004% of the expected K^+ molarity. The Rb dose used was also less than 0.2% of the average muscle Rb concentration of ten species of freshwater fishes (Sharif *et al.*, 1993).

After injecting the radionuclides, we withdrew 75- to 200-μL blood samples from the fish through the cannula at 10, 20, and 30 min and 1, 2, 4, and 10 h and 1, 1.5, and 2 d after injection. Most of the cannulae ceased functioning within 3 d after blood sampling began, and we removed them after anesthetizing the fish. All blood samples up to 3 d post-injection were from unanesthetized fish, but additional samples were taken at 4, 15, and 84 d after injection from the dorsal aorta of

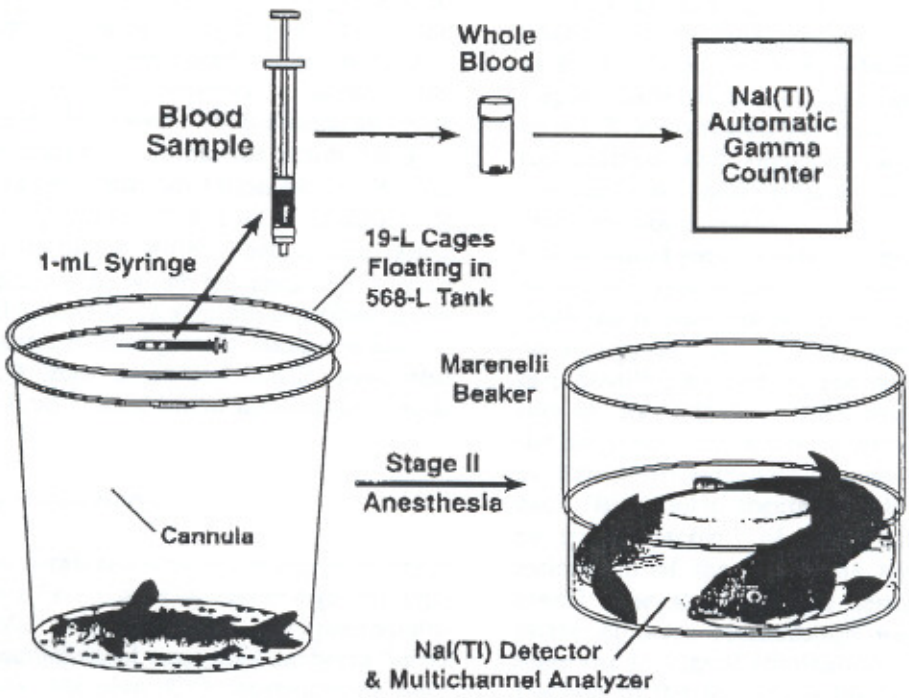

Figure 1. Diagrammatic representation of the regime for measurement of whole-body and blood kinetics of Rb and Cs in channel catfish. The cannulated fish were held in 19-L cages floating in 568-L aquaria. Intravascular injections of ^{86}Rb and ^{137}Cs were made through the cannulae, and blood samples were withdrawn through the cannulae at increasing intervals. Approximately once a week, the cages containing the fish were removed from the aquaria and immersed in an anesthetic solution. The whole-body burdens of the lightly anesthetized fish were then measured by placing each fish in a 4-L Marenelli beaker on a gamma spectroscopy system to measure whole-animal elimination of both metals.

attached to a 1-mL syringe. An equal volume of the modified Cortland saline was injected into the fish after each sample. We withheld food while the cannulae were in place, but resumed feeding the fish after removing the cannulae. However, the maintenance ration described above was reduced by one-third to compensate for the restricted activity of the caged fish. The cumulative volume of blood removed from each fish over a 12-wk period was less than or equal to 28% of the total blood volume initially present in the fish (estimated as 4% of wet body mass; Olson, 1992), with most of this removed during the last three samples (4, 15, and 84 d) after the fish resumed feeding. We counted the blood samples to a 1% 2-σ error or for 1 h in an automated 7.6-cm-wide by 8.3-cm-high well-type NaI(Tl) gamma counter (Packard Auto-gamma® 5530, Packard Instrument Company, Meriden, CT). Integrated sample and background counts were recorded within the 530–780 keV interval of the spectrum for the 662 keV ^{137}Cs photopeak, and within the 950–1200 keV interval for the 1078 keV ^{86}Rb photopeak. The integrated counts at 125 keV above and below both of the photopeak ranges were also recorded to determine whether radiations from either radionuclide were contributing to each other's photopeak. Radionuclide count yield of the blood samples for estimation of activity was determined by measurement of a standard of similar activity to the initial blood concentration of each radionuclide, which was prepared from an aliquot of the same stock solution described above for the injections. Background measurements of whole blood from unlabeled catfish were also made each time labeled blood samples were measured.

Whole-body measurements

Whole-body measurements of the fish were made for 180 s on a 7.6-cm wide by 7.6-cm high well-type NaI(Tl) solid scintillator detector/photomultiplier and multichannel analyzer (Canberra Series 85, Canberra, Meriden, CT) approximately once each week to measure the elimination of ^{86}Rb and ^{137}Cs. Each fish and its cage was removed from the tank and immersed in an aerated 19-L bucket containing approximately 15 L of 150 mg 2–3 min, until the fish's behavior (loss of ability to remain upright) indicated that stage II anesthesia had been attained. The fish was then removed from its cage and transferred to a covered 4-L polyethylene Marenelli beaker (GA-MA & Associates, Inc., Miami FL) containing 1 L of clean water (sufficient to cover the fish without allowing vertical movement). The beaker was then placed over the detector, with the curled body of the fish encircling the sensitive volume (Fig. 1).

Background count rates (counts s^{-1}) were determined by counting a water-filled container in the same geometry for the same length of time. Sample and background counts were recorded within the same intervals of the spectrum as described above for blood. The background count rate was subtracted from the gross count rate (the count rate of the labeled fish plus natural background) to obtain the net count rate (counts s^{-1}) for each individual. We corrected the net count rates for the amount of radionuclide lost due to physical decay since the beginning of each treatment by dividing them by $e^{-k_p t}$, where k_p is the fraction of the total radioactivity lost per unit time (defined as $(\ln 2)/T_p$, where t represents the elapsed time since the beginning of the experiment). The corrected net count rates were $\log_e$-transformed and regressed against time. The intercepts of these relationships were used to estimate count yield (counts/disintegration^{-1}), that is, count yield equals the initial corrected net count rate (estimated from the intercept of the whole-body elimination curve) divided by the total activity of ^{86}Rb or ^{137}Cs administered to the fish. Dividing each measured corrected net count rate by the individual fish's count yield converts the net count rate measurements into the amount of radioactivity of each radionuclide present in each fish (which, together with the information on specific activity of the injected dose, allows estimation of the remaining mass of each element). The slopes of the terminal elimination phase of these relationships were used to estimate the biological elimination rate constant (the fraction of the total body burden eliminated per unit time, k_b) and biological half-time ($T_b = (\ln 2)/k_b$). To determine whether the fish were cross-contaminating each other with their excreted radionuclides, we maintained one uncon-

taminated fish in the 20.0 °C tank for the duration of the experiment, and measured it on the gamma spectroscopy system each time to determine whether it had accumulated any ^{86}Rb or ^{137}Cs. Whole-body measurements of the fish that received injections of both metals were continued for 10 wk, after which point the ^{86}Rb declined through physical decay and biological elimination to levels that could no longer be measured accurately during a 180-s counting time (i.e., the SD of the net count rate increased to > 5% of the mean). After two more weeks, the fish were euthanized to determine ^{137}Cs tissue distributions and tissue to blood ratios.

Clearance-volume models of compartmental kinetics

The conceptual model used for estimating Rb and Cs compartmental kinetics is shown in Figure 2. We judged the appropriateness of this model in describing the data by means of methods described by Landlaw and DiStefano (1984), including testing the normality of the residual distributions, the coefficients of variation (CV) of the regression parameter estimates, the Akaike information criteria, performing *F*-tests of the multiple-partial correlation coefficients (R^2) of the functions (Kleinbaum *et al.*, 1988) and visual inspections of the fits (Boxenbaum *et al.*, 1974) and residuals. We also tested the residuals to determine whether serial samples from the same fish habitat exhibited significant positive or negative serial autocorrelation between measurement error and the amount of element present using the Durbin-Watson statistic.

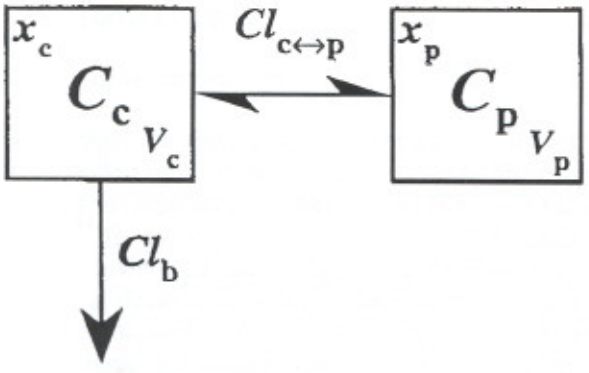

Figure 2. Conceptual model of intercompartmental distribution and whole-body clearance of Rb and Cs from channel catfish, showing the model parameters estimated for the clearance-volume model: V_c and V_p are the apparent volumes of distribution of the metals in the central and peripheral compartments, respectively; x_c and x_p are the masses of metal present in the central and peripheral compartments, respectively; C_c ($= x_c/V_c$) and C_p ($= x_p/V_p$) are the concentrations of the metals in the central and peripheral compartments, respectively; Cl_{c-p} is the intercompartmental clearance rate; and Cl_b is the total-body clearance rate.

We estimated the time-dependent concentration of each of the metals as:

$$\frac{dC_c}{dt} = \frac{Cl_{c-p}(C_p - C_c)}{V_c} - \frac{Cl_b C_c}{V_c}$$

$$\frac{dC_p}{dt} = \frac{Cl_{c-p}(C_c - C_p)}{V_p}$$

and

$$\frac{dx_f}{dt} = -Cl_b C_c,$$

where C_c and C_p are the central and peripheral compartments (mass volume^{-1}), respectively, V_c and V_p are the apparent volumes (mL g^{-1}) of the central and peripheral compartments, respectively, Cl_{c-p} is the intercompartmental clearance (mL d^{-1} g^{-1}) between V_c and V_p, Cl_b is the total body clearance (mL d^{-1} g^{-1}), and x_f is the total mass of each of the metals remaining in the body (also see Fig. 2). The blood and whole-body measurements were simultaneously fitted to these equations using NONLIN, a non-linear least squares statistical software package (Metzler, 1974) to estimate the values for V_c, V_p, Cl_{c-p}, and Cl_b. Initial parameter values for the model were obtained by curve-stripping the separate blood and whole-body data and then performing a preliminary fit of each fish's data set using a PC version of NONLIN to obtain the parameter estimates used in fitting the combined blood and whole-body data on the mainframe version of NONLIN. This preliminary modeling was repeated using a wide range of initial values, and yielded similar final estimates in each case. Weighting functions for the data were selected to accommodate the range of measured values. Because loss from both physical decay and biological elimination was high, we used a weighting function of $y_{(t)}^{-2}$, where $y_{(t)}$ was the measured value of

blood or whole body at each time (t), for the Rb data. For the less rapidly decreasing Cs data, we used a weighting function of $SDy_{(t)}^{-2} \times 100$, where $SDy_{(t)}$ was the standard deviation of the measured value of blood or whole-body at each time (t). At time $t = 0$, we assumed that x_f was equal to the dose, and that the concentration in the central compartment was equal to the mass of the dose divided by the apparent volume of the central compartment, i.e., $C_c = x_f / V_c$.

We estimated the total volume of distribution at steady state (V_{ss}, mL g^{-1}) as: $V_{ss} = V_c + V_p$, and calculated the elimination half-time predicted by the clearance-volume model ($t_{1/2,\beta}$, d) for each element as $t_{1/2,\beta} = \ln 2/\beta$, where

$$\beta = 0.5\left[Y - \sqrt{Y^2 - 4Z}\right],$$

$$Y = \frac{Cl_{c \leftrightarrow p} + Cl_b}{V_c} - \frac{Cl_{c \leftrightarrow p}}{V_p},$$

and

$$Z = \frac{Cl_{c \leftrightarrow p} Cl_b}{V_c V_p}$$

(modified from Stehly and Hayton, 1989). Other statistical tests were as described by Steel and Torrie (1980).

Tissue distributions

Nine of the catfish (3 females, 6 males) were killed by anesthetic overdose 12 wk after injection and dissected to determine the distribution of ^{137}Cs in organs and tissues (at this point, ^{86}Rb activity had decreased by a combination of physical decay and biological elimination to levels that could not be accurately determined for the smaller tissue and organ samples). Whole organs (liver, stomach, intestine, swim bladder, head and trunk kidneys, gonads, heart, and brain), visceral fat bodies, and samples of whole blood, bile, skin, gill filaments, red muscle (lateral line area), and white (axial myotomal) muscle from each individual fish were counted using similar methods as for blood. We estimated the percent of the remaining total body amount of ^{137}Cs that was present in each organ as the percent contribution of that organ's mass to the total body mass and determined the tissue to whole blood ratios. We assumed that the mass of the skin was 5% of body mass (... Schultz, unpublished data) and that the mass of blood was 4% of body mass (Olson, 1992). We also assumed that the mass of white muscle was 55% of body mass (Weatherley and Gill, 1987) and that the proportion of red to white muscle was the same as for rainbow trout, that is, 9% of white muscle by mass (Giblin and Massaro, 1973) or 5% of whole body mass.

Results

Ten fish ($n = 4$ at 20.0 °C and $n = 6$ at 27.5 °C) were used to compare Rb and Cs kinetics within individuals up to 84 d post-injection (70 d of whole body measurements). The fish fed readily after the cannulae were removed, and their body mass increased at 20.0 °C ($\bar{x}$ (SE): 11.8(7.38)%) and decreased at 27.5 °C ($\bar{x}$ (SE): −9.64(0.856)%).

Based on the low count rates of ^{86}Rb relative to ^{137}Cs, together with examinations of the spectrum intervals for the two radionuclides measured singly and in combination, both instruments appeared to have sufficient resolution to obviate the need for correcting for the contributions of each radionuclide to the other radionuclide's photopeak.

Whole-body elimination

For both metals, individual elimination curves estimated from whole-body count data alone were well described by single exponential models (Fig. 3A, B). The individual whole-body models were highly predictive for both Rb (all $r^2 \geq 0.98$) and Cs (all $r^2 \geq 0.92$). Coefficients of variation of the regression model intercepts (which were used to determine the count yield) were less than 1%. Rb T_b's estimated from the regression slopes averaged 16.4 d (95% confidence interval (CI): 14.7–18.1 d) at 20.0 °C and 15.8 d (95% CI: 14.0–17.6 d) at 27.5 °C. Cs T_b's were 5 to 7 times greater than those of Rb in the same individual fish and averaged 101 d (95% CI: 72.9–129 d) at 20.0 °C and 84.6 d (95% CI: 71.9–97.3 d) at 27.5 °C. The ratio of Cs T_b to Rb T_b was not affected by sex (Mann-Whitney U: $U = 2$, $U' = 16$, $P = 0.07$), but decreased with increasing temperature (Mann-Whitney U: $U = 2$, $U' = 22$, $P = 0.03$).

There was no relationship between initial body mass or the change in percent mass and the T_h of either element.

Clearance-volume modeling of compartmental kinetics

Of the models tested, a two-compartment clearance-volume model provided the best fit to both the Rb and Cs elimination data of the individual fish (Fig. 3A, B). The relative clearances and apparent volumes of distribution are depicted in Figure 4. The sizes of the Rb and Cs V_p's were both very large relative to the sizes for V_c's and the V_p's of both elements decreased by approximately 25% with increasing temperature. The overall V_{ss}'s (Fig. 4) for the two metals were not significantly different (Mann-Whitney $U: U = 73, U' = 87, P = 0.71$) and did not differ by temperature (Mann-Whitney $U: U = 53, U' = 115, P = 0.11$). The $Cl_{c \leftrightarrow p}$ values for the two elements differed dramatically (Mann-Whitney $U: U = 0, U' = 160, P < 0.0001$), indicating a nearly six-fold greater rate of Rb exchange between V_c and

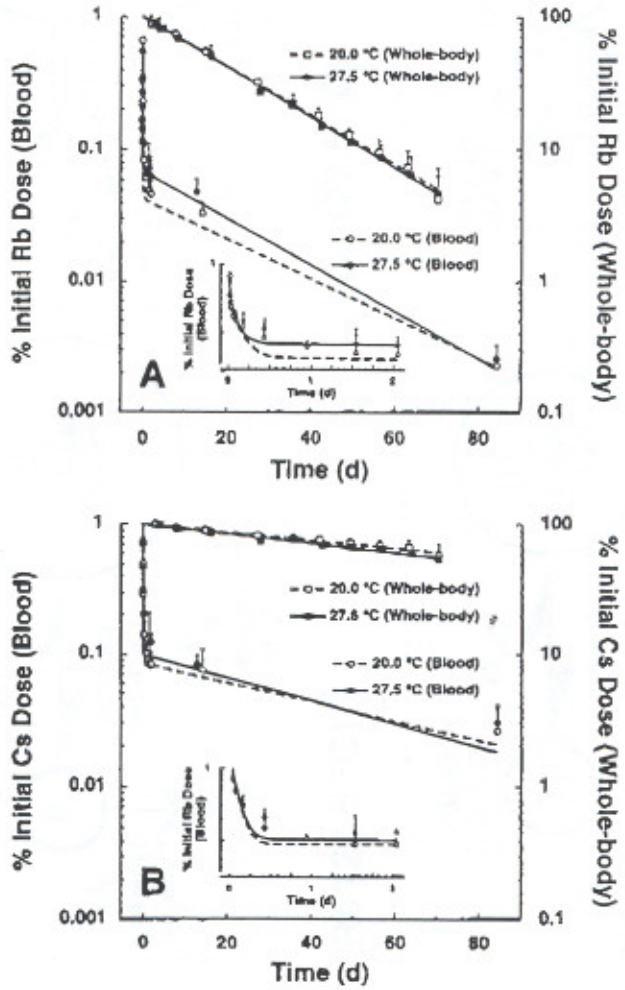

Figure 3. Whole-body and blood elimination of A) Rb and B) Cs from channel catfish at 20.0 °C (open symbols, $n = 4$) and 27.5 °C (solid symbols, $n = 6$). Whole-body measurements (squares) are of lightly anesthetized animals serially measured for 10 wk. Blood measurements (circles) are whole blood measurements serially measured for 12 wk. Inset figures show elemental kinetics in blood during the first 2 d after dosing. Data represent the remaining percent of the initial amount of each element administered to each fish (both stable carrier and radioactive tracer) in the blood or whole body, corrected for the physical decay of the radionuclides.

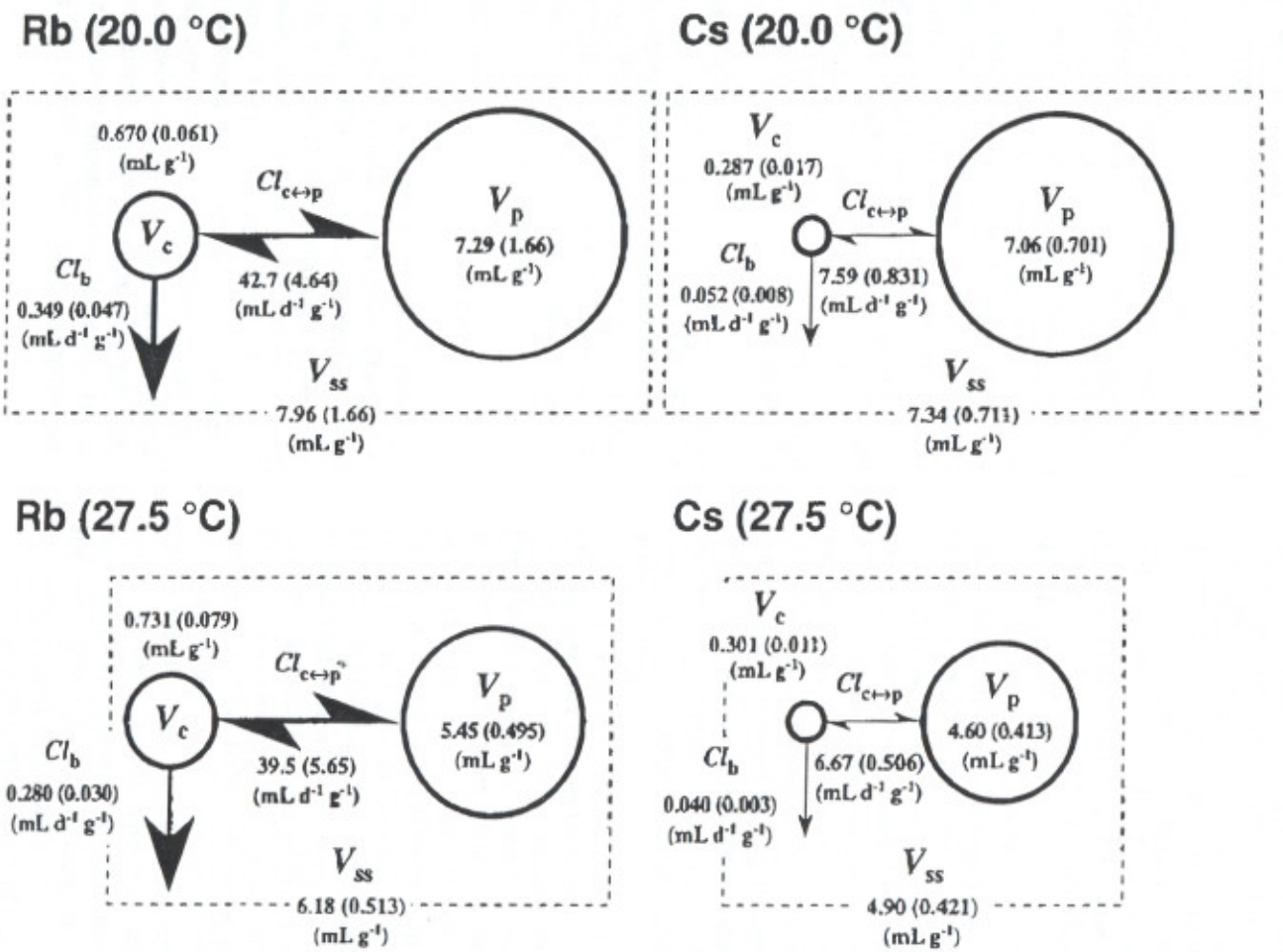

Figure 4. Pharmacokinetic parameters estimated from a clearance-volume model of combined whole-body and blood measurements of Rb and Cs after intravascular administration. Radionuclides of both metals were injected simultaneously into the dorsal aorta, and serial measurements of the amount of the remaining metals in blood and whole-body were simultaneously fitted to a two-compartment clearance volume model to estimate the model parameters for each fish. Parameters shown are: Cl_b (whole body clearance), $Cl_{c \leftrightarrow p}$ (intercompartmental clearance), V_c (apparent volume of the central compartment), V_p (apparent volume of the peripheral compartment), and V_{ss} (total apparent volume of distribution at steady state). The values shown represent the mean (and SE) of fish at 20.0 ($n = 4$) and 27.5 °C ($n = 6$).

V_p than for Cs (Fig. 4). The Cl_h's differed to the same degree (Mann-Whitney U: $U = 0, U' = 160, P < 0.0001$), with the Cl_h's of Rb 6.7 to 7.0 times greater than those of Cs at the same temperature (Fig. 4), indicating that the capacity of catfish to excrete Rb was about 7 times that of Cs.

The mean Cl_h's of both metals were about 1.3 times greater at the higher temperature (Fig. 4), but this difference was not statistically significant (Mann-Whitney U: Rb $U = 11, U' = 13, P = 0.83$; Cs $U = 10, U' = 14, P = 0.67$). However, Cl_h's of Rb were 7 times greater than those of Cs in the same fish (Fig. 4). The $t_{1/2,\beta}$'s calculated from the clearance-volume model parameters were nearly identical to the T_h's estimated from the whole-body measurements. Rb $t_{1/2,\beta}$'s averaged 15.5 d (95% CI: 10.6–20.6 d) at 20.0 °C and 15.6 d (95% CI: 13.9–17.0 d) at 27.5 °C. Cs $t_{1/2,\beta}$'s averaged 102 d (95% CI: 72.6–130 d) at 20.0 °C and 85.2 d (95% CI: 72.3–98.1 d) at 27.5 °C.

The mean Cl_{c-p} of both metals was slightly greater at the higher temperature (Fig. 4), but again, this difference was not statistically significant (Mann-Whitney U: Rb $U = 8, U' = 16, P = 0.39$; Cs $U = 10, U' = 14, P = 0.67$). There was no relationship between Cl_{c-p} values and either the T_h's or the $t_{1/2,\beta}$'s of either metal.

Tissue distributions and tissue/blood ratios

The organs, when corrected for their expected contributions to the total body mass (Table 2), contained nearly all of the Cs dose remaining in the fish. Over 85% of the remaining Cs was in white muscle, with significant quantities in red muscle, skin, and gill filaments (Table 2).

Tissue to blood Cs ratios were significantly greater than one for all well-perfused tissues (Table 2), and were highest in the spleen, white muscle, red muscle, heart, gonads, and gill filaments. The tissue to blood ratios decreased by an average of $20 \pm 4.0\%$ at the higher temperature (Table 2).

Discussion

Although ^{86}Rb has been frequently used as a metabolic tracer for K due to its longer physical half-time (18.7 d, compared with 12.4 h for ^{42}K), recent studies suggest that the kinetics of Rb in muscle and blood may not be identical to those of K (Dørup and Clausen, 1994). Thus, the clearances and volumes of distribution presented here for Rb in catfish may not reflect those of K. However, the T_h's observed for Rb resemble those of K more than they do Cs (Table 1), and the relative ratios of Cs:Rb T_h's that we observed in individual animals (5 to 7:1) were comparable to the ratio of Cs and K T_h's in bluegills (5 to 6:1, Kolehmainen, 1972).

In similar studies, fishes over similarly small sizes ranges have shown negligible size-dependent effects on their elimination of either element, compared with the influence of temperature (Peters, 1996; Peters and Newman, 1999). The effect of confinement on the movements of the catfish is also unlikely to have exerted much influence on elimination rates. A study of freshwater turtles showed no significant difference in Cs T_h's between turtles kept indoors in a controlled environment chamber, and others kept outdoors in an experimental pond (Peters and Brisbin, 1988).

The V_c's for Cs (Fig. 4) suggested that it was the approximate volume (about 30% of body mass) of the blood and well-perfused tissues, that is, liver, kidney, stomach, gonads, heart, brain, and gills. Our observations of tissue distribution and tissue to blood ratios also support this assumption, in that only a small fraction of the dose was found in these tissues, and because the Cs tissue to blood ratios were all less than half of the tissue to blood ratio of white muscle, which also constitutes the largest fraction of fish mass (Table 2). Tissue distributions of ^{137}Cs of chronically-contaminated yellow bullheads were similar to the acute distributions we observed (McCreedy et al., 1997). We observed no effect of temperature on the V_c of either metal. This was not unusual, as blood and well-perfused tissue volumes are generally not affected by acclimation temperature (Weatherley and Gill, 1987).

The large apparent volume of V_p and the high tissue to blood ratios of Cs present in red and white muscle suggested that these tissues were the primary contributors to this compartment. The decrease in the V_p of Cs with increasing temperature may be explained by decreased se-

Table 2. Organ distribution and tissue to blood ratios of ^{137}Cs in channel catfish (2 weeks after injection with a mean of 25.61(±0.48) μg kg^{-1} of ^{86}Rb and 588(±12) ng kg^{-1} of ^{137}Cs. The distribution of ^{137}Cs is relative to the total amount estimated to be present in the entire animal at that time, with means and standard errors back-calculated from arcsine-square root transformed percentage data. The tissue to blood ratios are the concentrations of ^{137}Cs in tissues (ng g^{-1}) divided by the concentration in whole blood (ng mL^{-1}) in the same fish

Tissue/organ	n for 20.0/27.5 °C	Percent of recovered Cs			Cs tissue to blood ratio		
		20.0 °C	27.5 °C	All	20.0 °C	27.5 °C	All
Blood	4/5	0.314 (0.058)	0.327 (0.015)	0.322 (0.025)	•	•	•
Bile	4/5	0.006 (< 0.001)	0.004 (< 0.001)	0.005 (0.001)	0.567 (0.076)	0.497 (0.067)	0.520 (0.049)
Liver	4/5	0.330 (0.050)	0.156 (0.039)	0.233 (0.042)	4.36 (0.938)	3.23 (0.791)	3.73 (0.600)
Spleen	4/5	0.056 (0.010)	0.069 (0.036)	0.063 (0.020)	9.74 (3.387)	9.17 (1.53)	9.45 (1.66)
Head kidney	4/5	0.101 (0.031)	0.035 (0.008)	0.064 (0.018)	4.92 (1.89)	4.31 (0.740)	4.58 (0.871)
Trunk kidney	4/5	0.340 (0.040)	0.140 (0.011)	0.229 (0.039)	7.37 (1.69)	5.58 (0.518)	6.37 (0.805)
Stomach	4/5	0.625 (0.028)	0.405 (0.028)	0.503 (0.043)	8.82 (1.55)	7.19 (0.452)	7.91 (0.732)
Intestine	4/5	0.686 (0.073)	0.354 (0.007)	0.502 (0.066)	6.38 (1.14)	5.89 (0.489)	6.11 (0.540)
Fat bodies	4/5	0.058 (0.012)	0.090 (0.025)	0.074 (0.014)	0.640 (0.120)	0.414 (0.108)	0.527 (0.086)
Swim bladder	4/5	0.075 (0.011)	0.059 (0.004)	0.066 (0.005)	2.08 (0.400)	1.75 (0.162)	1.90 (0.193)
Heart	4/5	0.106 (0.005)	0.066 (0.005)	0.084 (0.008)	13.4 (2.60)	10.1 (0.776)	11.6 (1.27)
Red muscle	4/5	6.21 (0.457)	6.24 (0.372)	6.23 (0.271)	17.6 (3.52)	15.5 (1.61)	16.5 (1.71)
White muscle	4/5	89.3 (1.04)	90.5 (0.358)	89.9 (0.509)	22.3 (3.06)	20.3 (0.966)	21.2 (1.40)
Skin	4/5	0.420 (0.132)	0.540 (0.146)	0.487 (0.096)	4.93 (1.68)	4.46 (0.607)	4.64 (0.763)
Brain	4/5	0.129 (0.007)	0.109 (0.013)	0.118 (0.008)	7.47 (1.91)	7.00 (0.460)	7.21 (0.819)
Testes	2/4	0.079 (< 0.001)	0.045 (0.004)	0.063 (0.006)	20.0 (NA)	6.21 (0.563)	8.97 (2.79)
Ovaries	2/1	0.803 (0.388)	0.532 (NA)	0.783 (0.328)	13.1 (3.37)	11.5 (NA)	12.6 (2.02)
Gill	4/5	1.02 (0.069)	0.822 (0.030)	1.03 (0.050)	5.30 (1.34)	3.72 (0.149)	4.32 (0.533)

questration in peripheral tissues and/or by increased sequestration in blood cells. The former process appears more likely to have occurred in the catfish, as the Cs tissue to blood ratios of most organs decreased with increasing temperature, indicating decreased sequestration. Moreover, the long component of the elimination of Cs from whole blood was more rapid than whole-body elimination, and was also constant with time (Fig. 3B), suggesting that there was no time-dependent accumulation of Cs in blood (i.e., within red blood cells).

Differences in the T_b's of different substances are due to the relative capacities for storage and/or excretion. Although the V_p's of both metals decreased with increasing temperature, the differences in Rb and Cs T_b's that we observed for catfish are apparently not due to different storage capacities, but to differences in excretory capacity (Cl_b). However, the Cl_b's of both metals

decreased with increasing temperature, which (other parameters being equal) should increase $t_{1/2,\beta}$. This suggests that the observed decrease in Cs T_b with increasing temperature may be due to changes in the amount of peripheral storage, rather than changes in excretory capacity. If this is true, then one might expect that cold-water fishes might accumulate more radiocesium than warm-water species. This may help to explain why the fish/prey concentration ratios observed in Scandinavian lake fishes (Carlsson, 1978; Forseth et al., 1991) tend to be about 2 to 3 times higher than for warm-water lake species (Kohlehmainen, 1972; Whicker et al., 1990).

The large $Cl_{c \leftrightarrow p}$ values we obtained for Rb (Fig. 4) suggested that blood flow to V_p was the predominant process influencing intercompartmental clearance of this metal. The cardiac output rate of 419–990 g channel catfish was estimated to be 2.4 L h^{-1} kg^{-1} fish (58 mL d^{-1} g^{-1}) at 21°C (McKim et al., 1994), and the blood flow to muscle has been estimated for several teleosts as approximately 40% of the cardiac output (Kolok et al. 1993): 0.4×58 mL d^{-1} g^{-1} or 23 mL d^{-1} g^{-1}. Our 95% confidence intervals for $Cl_{c \leftrightarrow p}$'s (29 to 56 mL d^{-1} g^{-1} at 20.0°C, 28 to 51 mL d^{-1} g^{-1} at 27.5°C) are similar to this value. The much lower $Cl_{c \leftrightarrow p}$ of Cs may reflect differences in its diffusion rate and/or its relative affinity compared with Rb for binding to cell membrane proteins (e.g., Na^+-K^+ ATPases).

All animals accumulate high concentrations of K, Rb, and Cs in the intracellular portions of soft tissues, but both Rb to K and Cs to K tissue concentration ratios are much greater than would be expected from the relative abundance of these elements (9100:32:1, K:Rb:Cs) in the environment. However, the tissue concentration patterns of Rb and Cs differ substantially from each other. Like K, Rb is not accumulated in any specific organ or tissue (Underwood, 1958; 1971). In the case of Cs, however, significant quantities are expected to concentrate in skeletal muscle relative to other tissues (Narayanyan and Eapen, 1971; Koulikov and Ryabov, 1992; McCreedy et al., 1997). Thus, once Rb and Cs ions have entered muscle cells via active transport, their passage back across the membrane may reflect differences in passive diffusion rates. The high tissue muscle (Table 2) are consistent with this assumption.

Application of pharmacokinetic techniques to traditional radionuclide kinetics studies should improve predictions of organismal radionuclide kinetics in the environment. Intravascular cannulation permits the delivery of quantifiable amounts of radionuclides directly into the bloodstream, which appears to reduce the difficulties resulting from time lags in absorption and distribution characteristic of oral gavage techniques. This technique can also be applied to obtain better information on environmental effects on ^{137}Cs assimilation and uptake. Future studies of the kinetics of radiocesium and other radionuclides should consider the influence of other factors (e.g., age, sex) on both compartmental kinetics and tissue distributions. Such information, if combined with information on the physical properties of contaminant ions and their effects on enzyme kinetics, may lead to more general predictions on the potential for uptake and accumulation of these substances by organisms in contaminated environments.

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