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Selenium levels in selected species of aquatic birds on Imperial National Wildlife Refuge

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SELENIUM LEVELS IN SELECTED SPECIES OF AQUATIC BIRDS ON IMPERIAL NATIONAL WILDLIFE REFUGE

by

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ABSTRACT

Five species of waterbirds were collected from five sites on Imperial National Wildlife Refuge between April and August of 1993. There were previous baseline contaminants data for all sites. Sites were of two distinct habitat types. Backwater lakes have a direct connection to the mainstem of the Colorado River, and seep lakes receive river water only via seepage through the soil column. Selenium concentrations in liver, kidney, and muscle tissues were consistently higher in birds collected from backwater lakes than those collected from seep lakes. Eighty-one percent of the birds collected on backwater lakes ($n=52$) were above the effect threshold for reproductive impairment or embryotoxicity (10 ppm dry weight in livers). Herbivorous birds had significantly ($p \leq 0.05$) lower tissue selenium levels than those species feeding on animal matter. Of the birds feeding on fish and invertebrates, 83% ($n=47$) had selenium levels in liver above the effect threshold. Differences in selenium concentrations based on diet suggest food chain cycling of selenium. Eggs from waterbirds as well as those from neotropical migrants were above the 3 ppm embryotoxicity threshold.

INTRODUCTION

In the 1930's, researchers observed the first toxic effects of selenium in livestock; the disease known as "blind staggers disease" or "alkali disease." In contrast, hazards to wildlife associated with elevated selenium concentrations were first documented much later at Kesterson National Wildlife Refuge (Kesterson NWR), California. In 1983 elevated selenium levels in irrigation sub-surface drain water caused adult mortality, reproductive failure, embryonic mortality and developmental abnormalities in many aquatic bird species nesting on the refuge (Ohlendorf et al., 1986a, 1986b, 1987, 1988, Presser and Ohlendorf 1987). The impacts on the nesting and migratory birds at Kesterson NWR prompted evaluation of selenium levels in the biota from other wildlife refuges in arid and semi-arid climates (Saiki 1987).

Imperial National Wildlife Refuge (Imperial NWR) encompasses 30 miles of the lower Colorado River (from Davis Dam south to the international boundary, Figure 1) along the Arizona-California border approximately 65 miles north of the United States-Mexico international border. Imperial NWR is used extensively by migrating and wintering waterfowl and shorebirds, and is critical foraging and nesting habitat for various species of waterbirds and marsh birds including the federally listed endangered Yuma clapper rail (*Rallus longirostris yumanensis*). The average rainfall is 5-10 cm with an average summer high temperature of 40°C and an average low of 21°C (Ohmart et al. 1988). The

refuge fits the profile for high selenium risk because of these semi-arid conditions (high evaporation rate) and high natural soil alkalinity (Riley and Riley 1979). Previous data indicated that marsh birds in the lower Colorado River valley were at moderate to high risk of selenium-induced teratogenicity (Rusk 1991). In addition, recent baseline data from Imperial NWR have shown elevated selenium levels in sediment, biota, invertebrates, and fish (Lusk 1993).

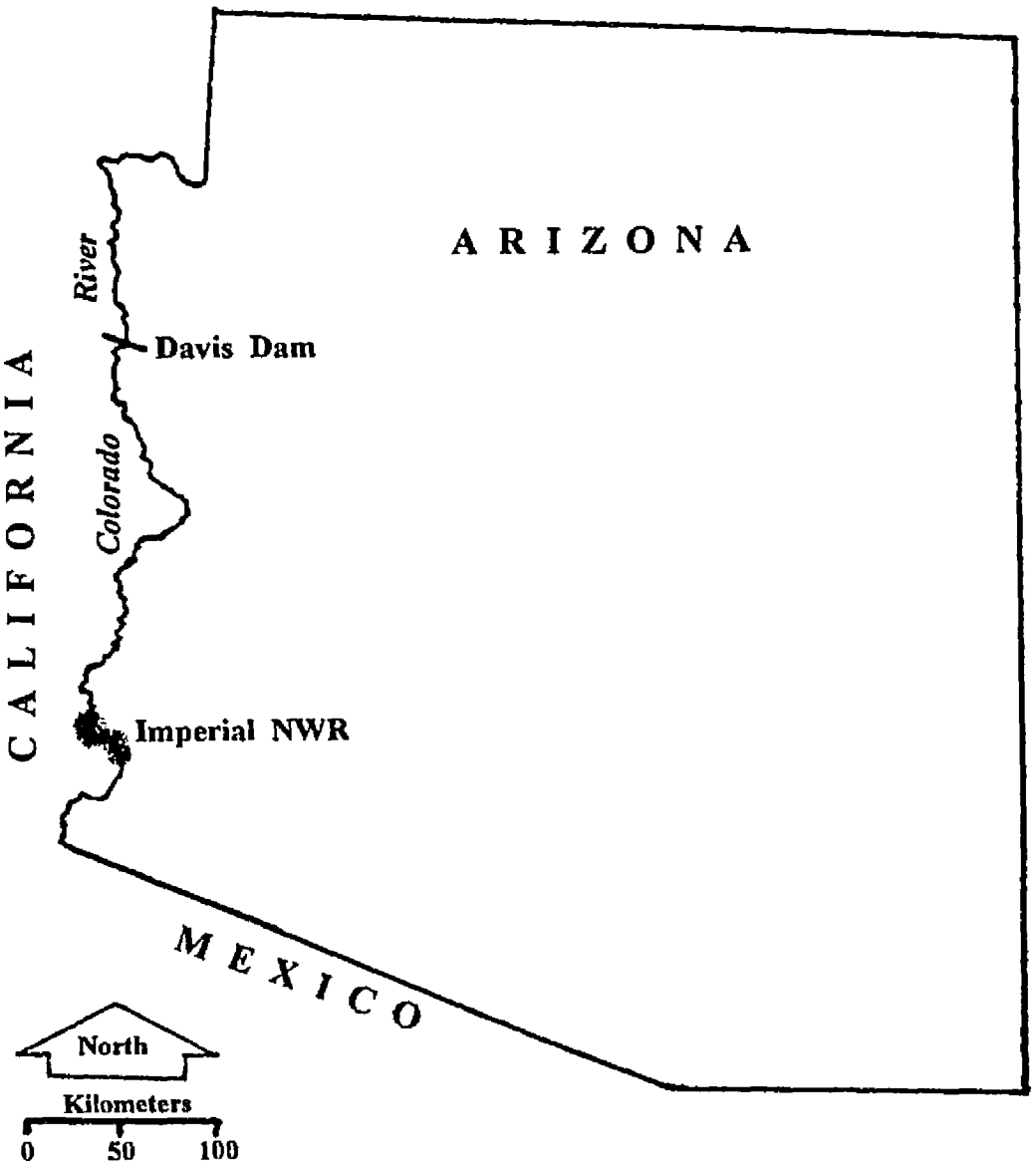


Figure 1. The location of Imperial National Wildlife Refuge relative to state borders and international boundaries.

BACKGROUND

Study Area:

Historical selenium levels for biota from the lower Colorado River are not well documented. The U.S. Fish and Wildlife Service monitored contaminant levels in fish from 1972 to 1985 as part of the National Contaminant Biomonitoring Program (NCBP) (Walsh et al. 1977, Lowe et al. 1985, Schmitt and Brumbaugh 1990), but tissues were not always analyzed for selenium (May and McKinney 1981). The data that are present have shown that selenium levels in fish tissues from the lower Colorado River were consistently the highest in the nation. Dissolved selenium concentrations in the lower Colorado River have been above the national baseline since 1975, when the United States Geological Survey began monitoring (Radtke et al. 1988). Selenium concentrations in bottom sediments of the lower Colorado River have consistently been above the baseline concentrations for western soils (King et al. 1993, Radtke et al. 1988). Baseline values are defined according to geometric means for similar soils in the western United States and waters across the nation. Levels in the lower Colorado River have consistently been above the 95 percent level for soils and 75 percent level for surface waters.

Recent data from Imperial NWR have consistently indicated elevated (above background) selenium levels in water, sediment, and biological samples (Radtke et al. 1988, Rusk 1991, Bell-McCaulou 1993, King et al. 1993, Lusk 1993).

However, the source of elevated selenium levels is not the lower Colorado River (Lowe et al. 1985). Instead, dissolved selenium in the water column is derived from several sources in the upper Colorado River basin, including natural weathering of seleniferous soils or rocks including mine tailings of various seleniferous ore deposits, combustion of seleniferous coal at electric generating stations, and irrigation return flows from areas with seleniferous soils (Radtke et al. 1988, Welsh and Maughan 1994). Thus, the major sources of selenium to fish and wildlife are from anthropogenic activities (including fossil fuel combustion) and runoff from seleniferous areas (Eisler 1985). These observations would suggest that selenium levels in the Colorado River may historically have been higher than the expected "normal" background levels.

The upper Colorado River basin includes the Green River basin in Utah, the Gunnison River basin in Colorado, and the San Juan River basin in New Mexico. Dissolved selenium in the middle Green River is <1 ppb as it enters northern Utah but increases to 2 ppb before it enters the Colorado River (Stephens et al. 1992). In the Gunnison River, selenium levels are 8 ppb above its confluence with the Colorado River (Butler et al. 1991). Selenium concentrations in the water of the San Juan River and its tributaries are 2 ppb (Blanchard et al. 1993). Waterborne selenium concentrations of 2 ppb or greater have high potential for food-chain bioaccumulation (Peterson and Nebeker 1992) and reproductive effects, and have been considered hazardous to the health and

long-term survival of fish and wildlife populations (Lemly 1993). In fact, there are elevated selenium levels in the birds collected from all three drainages. Selenium levels in aquatic bird tissue ranged from 1.8 to 108 ppm dry weight along the middle Green River (Stephens et al. 1992), 6.5 to 54.2 ppm dry weight in waterbird livers along the lower Gunnison River (Butler et al. 1991) and from 2.7 to 103 ppm dry weight in avian liver and kidney tissues collected along the San Juan River and its tributaries (Blanchard et al. 1993). The toxicity guideline for selenium in the liver tissues of aquatic birds is 10 ppm dry weight (Lemly 1993, Ohlendorf et al. 1993). Above 10 ppm dry weight there are significant dangers of teratogenic and toxic effects to avian species.

Habitat Differences:

Lusk (1993) reported that selenium levels were elevated in biota from backwater lakes (Adobe, Island, and Bee lakes) but not in those from seep lakes (McAllister and Butler lakes); backwater lakes are those directly connected to the river by one or more canals whereas seep lakes receive water from the river only via seepage. Lusk (1993) hypothesized that seep lakes receive lower levels or different forms of selenium than backwater lakes because of filtration through the soil column. In addition, the water quality conditions (i.e., high salt content, pH levels, sulfate concentrations, conductivity and hypoxia) in seep lakes may further act to reduce selenium levels. Differences in selenium levels in the biota from

different habitats are maintained by restricted access and egress to and from seep lakes. Juvenile birds (flightless) should also be restricted in their access to and egress from some habitats but adult birds are not similarly restricted; adults have equal access to the river, backwater lakes and seep lakes. If adult birds move between areas, one would not expect close correlations between tissue selenium levels and specific environments (Lusk 1993). However, if adult birds feed exclusively in one type of habitat, one should find such correlations. Therefore, the first objective of this study was to determine whether there was a relationship between selenium levels in the tissues of adult and juvenile birds and the habitats in which they were seen feeding. Specifically, a comparison was made between selenium levels in adult birds seen feeding in seep lakes versus those seen feeding in backwater lakes. Comparable levels in all adult birds regardless of feeding habitats would indicate all birds were equally exposed to selenium. This same comparison was also made for two feeding guilds of juvenile (flightless) birds. Ohlendorf et al. (1990) hypothesized that juvenile birds reared at a specific site usually provide the best indication of site-specific selenium bioaccumulation (the propensity of aquatic organisms to biologically amass certain elements to concentrations greater than the levels in the food consumed) . These data should indicate if birds fledged on backwater lakes are at greater risk to selenium toxicosis than those fledged on seep lakes.

Dietary Differences:

Selenium levels in the tissues of birds from Kesterson NWR were similar whether they fed on animal material or plant material (Ohlendorf et al., 1986a, 1986b, 1987, 1988). However, data from prey items on refuges along the lower Colorado River have indicated higher levels occur in animal matter than plant material (Lusk 1993). Therefore, this study also tested whether there were differences in selenium levels between birds that eat fish and invertebrates and those that eat plants. A detritus/detritivore mediated uptake mechanism has been suggested as the pathway of selenium accumulation in invertebrates in this system (Lusk 1993). Others have also suggested that recycling of organic selenium contained in decaying plant and animal detritus and its associated microbial elements may be responsible for high selenium residues in the food-chain and consumer species (Lemly and Smith 1987, Saiki and Lowe 1987, Saiki et al. 1993).

Selenium Bioaccumulation:

Selenium is concentrated in the tissues when it is taken up by organisms faster than it can be excreted and in excess of metabolic needs. It is incorporated in small amounts into the animal enzyme glutathione peroxidase, which protects the cell from damage from molecular oxygen, as well as into several cytochromes, myoglobulin, and hemoglobin (Eisler 1985). Excess selenium may be

incorporated into amino acids and proteins by methylation (Eisler 1985, Goyer 1993). Some authors have suggested that bioaccumulation from food items results in higher toxicity than that from aqueous exposure (Hamilton et al. 1990, Saiki 1990, Skorupa and Ohlendorf 1991, Besser et al. 1993, Coyle et al. 1993). Selenium levels in dietary items of aquatic birds are greatly increased compared to those in water due to the bioaccumulation of selenium through the aquatic food chain. Therefore, fish and birds that consume prey that have high selenium levels in their tissues are exposed to the greatest risks of bioaccumulation (Lemly and Smith 1987, Saiki and Lowe 1987, Hothem and Ohlendorf 1989).

Tissue Differences:

Different bird tissues accumulate selenium at different rates. For example, selenium accumulates much more rapidly in the liver than in breast muscle (Fairbrother and Fowles 1990). In the laboratory, selenium concentrations in the liver were comparable to those in the diet (10 ppm selenomethionine) eight days after the onset of exposure. In contrast, it took 81 days for breast muscle to reflect dietary selenium concentrations (Heinz et al. 1990). Selenium elimination rates also follow different patterns in different tissues. In laboratory tests, selenium is lost at a faster rate from the liver than from the muscle; half-times are 18.7 and 30.1 days, respectively (Heinz et al. 1990). These differences may arise because the liver is an organ of active metabolism and excretion, whereas the

muscle incorporates selenomethionine in the place of methionine in the synthesis of proteins (Beilstein and Whanger 1987). If the laboratory data are applicable to field situations and if adult birds feed in only one habitat type (i.e., seep or backwater lake), the ratio of selenium in the muscle to that in the liver might be used as an indicator of residence time in an affected area. Furthermore, we may be able to use tissue levels to estimate the onset and duration of exposure. If selenium concentrations in breast muscle and liver are both comparable to background concentrations, then exposure is long term and must be considered chronic. In contrast, if selenium concentrations are comparable to background levels in liver but not in breast muscle, then exposure time must be short term or acute. If selenium concentrations are high in breast muscle but are low in liver, then selenium exposure occurred at some earlier time and is being eliminated from the body.

The kidney is the primary route by which selenium is excreted from the body. Therefore, this organ typically exhibits high concentrations of selenium when exposure has occurred (Fairbrother and Fowles 1990). Ratios of selenium in kidney and liver have been used to correct for toxicity differences among species and diets (Ohlendorf et al. 1990). Dietary selenium levels are assumed to be "normal", when kidney:liver ratios are greater than 1, and elevated when they are less than 1 (Ohlendorf et. al. 1990, Ohlendorf and Skorupa 1989). In addition, when selenium content in kidneys is elevated above that in liver, dietary

selenium levels are assumed to be low. In contrast, when levels in the liver equal or exceed those in the kidney, selenium intake is assumed to be high (Goede 1985). Therefore, another objective of this study was to compare liver/kidney/muscle ratios and to assess patterns of selenium exposure.

Dietary selenium levels are reflected in eggs of aquatic birds. Elevated amounts of selenium in the diet have been associated with decreased egg hatchability in laboratory studies of mallards (*Anas platyrhynchos*) and domestic chickens (*Gallus gallus*) (Heinz and Fitzgerald 1993, Heinz et al. 1987, Ort and Latshaw 1978, Arnold et al. 1973) and in free ranging aquatic birds at Kesterson NWR (Ohlendorf et al. 1986b). Selenium is rapidly accumulated in eggs (about two weeks), therefore a female would only have to feed in a high selenium environment for a short time before selenium concentrations reached equilibrium in the egg (Heinz 1993). The threshold level at which teratogenic or toxic effects may occur in aquatic bird eggs is 3 ppm dry weight (Lemly 1993). Thus, the final objective of this study was to determine selenium content in eggs and to relate these levels to potential effects on birds.

METHODS

Sites Sampled and Species Collected:

Samples were collected from three backwater lakes (Adobe, Island, and Bee) and two seep lakes (Butler and McAllister) (Figure 2). Species were selected for collection based on availability across sites, as well as foraging behavior. Eggs, young (chick, juvenile) and adults of five species of marsh birds were collected. Species collected included: American coot, *Fulica americana*, a local common breeder and abundant winter resident throughout the lower Colorado River Valley; common moorhen, *Gallinula chloropus*, a fairly common breeder and local permanent resident; least bittern, *Ixobrychus exilis*, a common breeder from April to September; green heron, *Butorides virescens*, a fairly common summer resident and breeder from March through September; and pied-billed grebe, *Podilymbus podiceps*, a common resident and breeder; (Rosenburg et al. 1991). Coots and moorhens were classified as herbivores (Mulholland and Percival 1982, Eley and Harris 1976). Bitterns and herons were classified as fish- and invertebrate-feeders based on information from the literature (Ehrlich et al. 1988) and confirmed by stomach content analysis.

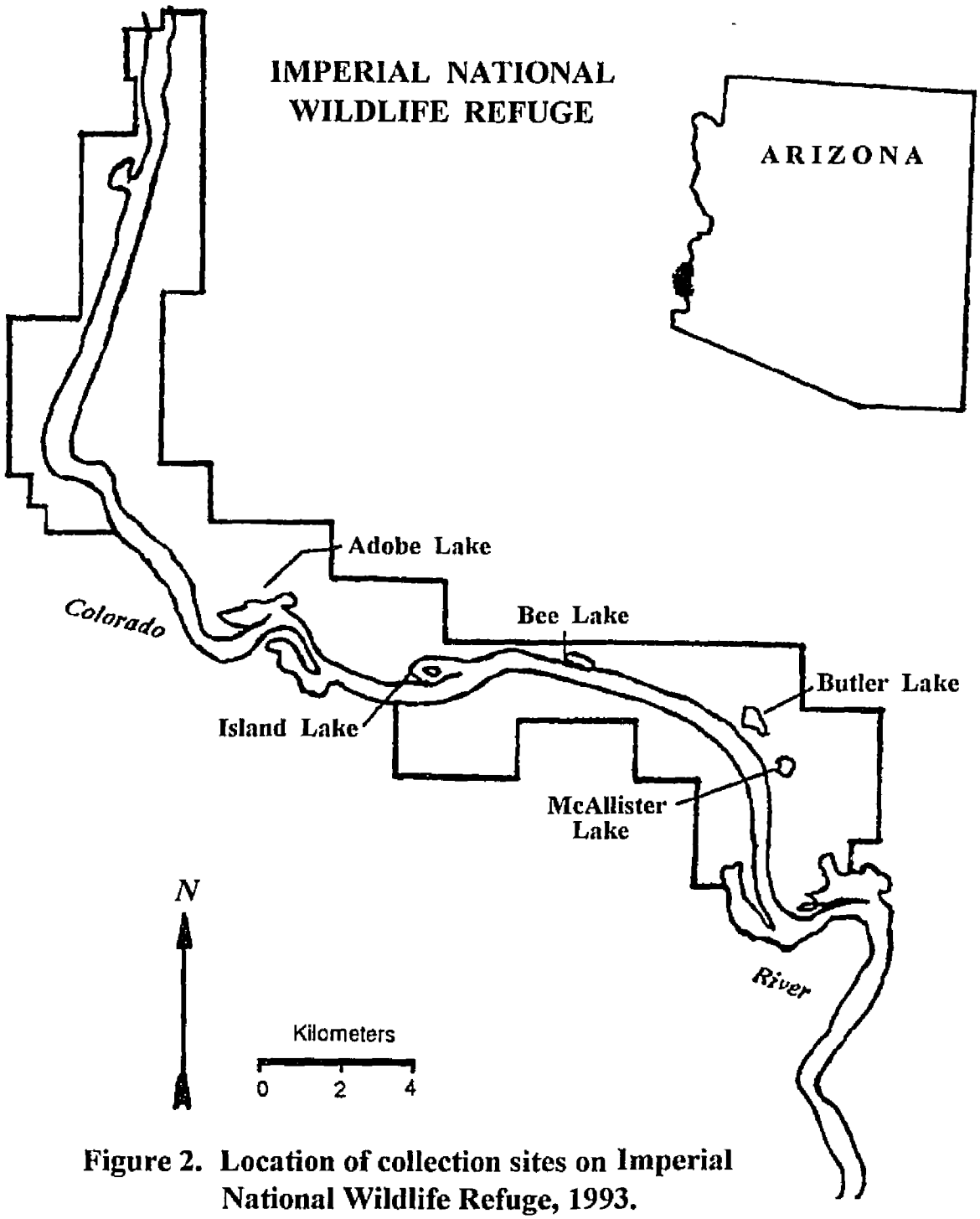


Figure 2. Location of collection sites on Imperial National Wildlife Refuge, 1993.

Collection and Preparation:

Collections were made during the breeding season between April and August 1993. A 20-gauge shotgun loaded with number 6 steel shot was used to collect birds. Specimens were placed on wet ice and transported to refuge headquarters for dissection. Dissection took place within a few hours of collection. Liver, kidney, and breast muscle were dissected from each bird using a stainless steel scalpel. Egg masses were extracted when present. Tissues were weighed and placed in a labelled, sterile Whirlpak bag and frozen in a commercial freezer. Dissection equipment was rinsed with deionized water before each dissection. Dissections were performed on aluminum foil and disposable, sterile, latex gloves were worn during each procedure. Eggs were measured and processed in accordance with the guidelines of the Patuxent Analytical Control Facility (PACF) (U.S. Fish and Wildlife Service 1990). They were then placed in a chemically cleaned jar and frozen. Samples were transported frozen on wet ice from refuge headquarters to the University of Arizona where they were placed in a walk-in freezer until shipped on dry ice to a PACF contract laboratory. Prey items were collected from the gastrointestinal tract of carnivorous birds when present. The number and species of prey were used in estimating residence time.

Selenium Analysis:

Selenium analyses were conducted by Research Triangle Institute (RTI), Research Triangle Park, North Carolina. RTI is a contract laboratory of the PACF. Selenium was analyzed using Graphite Furnace Atomic Absorption (GFAA). Tissue samples were homogenized using a food processor. A portion of the tissue sample was then freeze-dried for determination of moisture content and ground to 100 mesh with a mill. Digestion for GFAA measurement was conducted using a CEM microwave oven. From 0.25 to 0.5 g of freeze-dried sample was heated in a capped 120 ml Teflon vessel in the presence of 5 ml of Baker Instra-Analyzed nitric acid for three minutes at 120 watts, three minutes at 300 watts, and fifteen minutes at 450 watts. The residue was then diluted to 50 ml with laboratory pure water. GFAA measurements were made using a Perkin-Elmer Zeeman 3030 or 4100ZL atomic absorption spectrometer.

Quality Assurance/Quality Control:

Quality assurance used by RTI for the chemical determinations included the analysis of duplicate samples, procedural blanks, analysis of standard reference materials (i.e., dogfish liver [NRCC DOLT-2] and lobster hepatopancreas [NRCC TORT-1]), and spike recoveries. PACF approved all quality assurance/quality control samples on all analyses.

Statistical Analysis:

Selenium concentrations were log-transformed prior to statistical analysis to normalize the variance (Note: Geometric means are reported rather than arithmetic means because they are less influenced by extreme or "outlier" values, and hence provide a more conservative estimate of central tendency {Saiki and Palawski 1990}). Range and standard error of the geometric mean are reported as measures of dispersion. Bartlett's test was used to determine if variances were equivalent. Statistical analyses were performed using SAS (SAS Institute Inc. 1987). Analysis of variance (ANOVA) employing the GLM procedure with a significance level of $p \leq 0.05$ was used to determine differences in mean tissue selenium concentrations between species, sites, and site classes. Significant differences were further analyzed with Tukey's test to determine the exact location of significant differences. After among species comparisons were made, species were grouped into classes according to food habits (herbivores or carnivores) and compared using linear contrasts. Linear contrasts allowed tissues to be grouped by species and compared, but did not allow species with high tissue concentrations of selenium but low sample numbers to bias the analysis.

Kidney:liver ratios were used to assess dietary selenium intake. Pearson's correlation coefficient was used to measure the relationship between tissue levels of selenium in three species. Plots were used to illustrate trends based on selenium levels in liver over time (the date of collection in julian days). The date

of collection was used to determine trends in selenium levels between individuals collected at the beginning of the breeding season in April, and those collected later in the season in August.

Selenium levels in liver and muscle were compared to selenium levels in food items for the specific habitat in which the bird was collected. Residence time, and therefore exposure to selenium, were determined based upon selenium concentrations in liver and muscle when compared to selenium levels in prey species. Accumulation and elimination rates (Heinz 1990) were used to develop models of exposure time for adult birds taken from seep lakes and backwater lakes.

RESULTS

There were no significant differences in selenium concentrations between adult male and female birds for any of the tissues or species (Table 1). Therefore, all subsequent analyses combined the data from both sexes. Levels are reported on a dry weight basis unless otherwise stated; percent moisture content for each sample is listed in Appendix A. There were insufficient numbers of eggs or young birds (chicks or juveniles) for statistical analyses. However, data from all other matrices were analyzed statistically. Despite the small sample size, pied-billed grebes had the highest levels of selenium in their tissues, therefore, were included in statistical analyses. Selenium concentrations were analyzed to the lowest limit of detection (ppm, dry weight) ≤ 1.2255 in liver, ≤ 5.102 in kidney, ≤ 0.644 in muscle, and ≤ 0.7716 in eggs.

Table 1. Geometric means of selenium concentrations (ppm, dry weight) in adult tissues by species and gender (n = number of birds analyzed). Analysis of variance for gender at each site for each species ($p \leq 0.05$).

			Liver		Kidney		Muscle	
Species	♂ (n)	♀ (n)	Male	Female	Male	Female	Male	Female
American coot	10	5	6.7	6.3	12.0	12.4	3.9	3.5
Common moorhen	11	6	9.4	9.3	12.7	11.6	4.0	2.6
Least bittern	8	19	17.1	14.1	31.9	26.1	4.6	3.2
Green heron	5	5	16.2	15.8	24.7	20.2	4.5	4.6
Pied-billed grebe	0	3	---	26.1	---	25.5	---	10.2
Totals or averages	34	38	12.4	14.3	20.3	19.2	4.3	4.8
F-Value			$F = 0.09$		$F = 0.79$		$F = 0.24$	

Temporal Trends:

Bird collections on seep lakes were completed during the early breeding season. Tissue concentrations in these birds were lower than those of birds also collected during the early breeding season from backwater lakes (Table 2). There were no apparent differences in selenium levels between birds collected during the early and late breeding seasons on backwater lakes for any species (Table 3).

Table 2. Comparison of geometric means of selenium concentrations (ppm, dry weight) in livers of adult birds collected during the early breeding season on backwater lakes versus those collected on seep lakes (n = number of birds analyzed, SE = standard error).

	Backwater Lakes			Seep Lakes		
Species	Early Season	SE	(n)	Early Season	SE	(n)
American coot	9.46	1.18	3	5.25	1.09	9
Common moorhen	14.29	1.03	8	6.06	1.09	8
Least bittern	21.26	1.08	9	10.45	1.11	14
Green heron	18.02	1.17	4	12.62	1.09	3
Pied-billed grebe	25.1	1.11	2	22.50	---	1

Table 3. Comparison of geometric means of selenium concentrations (ppm, dry weight) in livers of adult birds collected on backwater lakes in early breeding season versus those collected in late breeding season (n = number of birds analyzed, SE = standard error).

	Backwater Lakes					
Species	Early Season	SE	(n)	Late Season	Se	(n)
American coot	9.46	1.18	3	8.85	1.03	3
Common moorhen	14.29	1.03	8	9.97	---	1
Least bittern	21.26	1.08	9	23.32	1.22	4
Green heron	18.02	1.17	4	17.27	1.17	3
Pied-billed grebe	25.1	1.11	2	28.50	---	1

Site Comparisons:

There were significant differences between selenium levels in liver, kidney, and muscle among birds at individual sites (Table 4). Birds from Butler Lake (seep lake) had the lowest geometric mean selenium levels in all tissues. The highest selenium concentrations in liver and kidney were detected in samples collected from Bee Lake (backwater lake). Selenium concentrations in muscle tissue were highest at Adobe Lake (backwater lake).

Table 4. Geometric means (GM) ± standard error (SE) and ranges of selenium concentrations (ppm, dry weight) in adult birds for all species by site and tissue. Means with different capital letters are significantly different ($p \leq 0.05$) among tissues (n = number of birds analyzed).

Site	n	Liver GM±SE	Range	Kidney GM±SE	Range	Muscle GM±SE	Range
Adobe Lake	13	15.02 A ±1.09	9.4-28.5	27.18 A ±1.14	11.2-53.3	6.36 A ±1.09	3.9-13.2
Island Lake	18	16.17 A ±1.11	7.3-34.2	21.30 A ±1.19	4.4-60.9	5.71 A ±1.06	3.6-8.1
Bee Lake	7	20.11 A ±1.13	14.3-35.6	22.35 A ±1.10	15.8-31.9	5.87 A ±1.08	3.8-7.0
McAllister Lake	17	8.87 B ±1.10	4.3-21.1	16.69 B ±1.19	8.3-63.2	2.57 B ±1.15	0.7-7.5
Butler Lake	17	6.85 B ±1.12	3.4-18.0	12.59 B ±1.11	6.1-23.0	2.08 B ±1.11	1.2-4.7
F-Value		$F = 24.25$		$F = 4.37$		$F = 20.79$	

Site class comparisons revealed differences between backwater lakes and seep lakes. Geometric mean selenium levels were significantly lower ($p \leq 0.05$) in all tissues (i.e. liver, kidney, and muscle) of birds collected from seep lakes (Butler and McAllister) than in birds from backwater lakes (Adobe, Bee, and Island) (Table 5). For example, selenium levels in muscle tissue were significantly higher ($p \leq 0.05$) in moorhens and bitterns collected from Bee, Island, and Adobe lakes (backwater lakes) than in those from McAllister and Butler lakes (seep lakes).

Table 5. Geometric mean selenium concentrations (ppm, dry weight) ± standard error averaged over all species and tissues at each site class. Means with different capital letters are significantly different ($p \leq 0.05$) among tissues (number in parentheses = number of birds analyzed).

Tissue	Seep lake			Backwater lake			*F*-Value
Liver	7.79 ± 1.08	A	(34)	14.8 ± 1.01	B	(38)	$F = 27.35$
Kidney	14.43 ± 1.10	A	(34)	20.8 ± 1.02	B	(38)	$F = 7.36$
Muscle	2.32 ± 1.09	A	(34)	5.7 ± 1.04	B	(38)	$F = 16.10$

Species Comparisons:

There were statistical differences in selenium levels between species and among and between sites. Coots had the lowest levels of selenium in liver and kidney (Table 6). In addition, selenium levels in the livers and kidneys of coots and moorhens were significantly lower ($p \leq 0.05$) than those in herons and bitterns. There were statistical differences (linear contrasts, $p \leq 0.05$) between selenium levels in liver and kidney of plant eating species and species that feed on animal matter. Herbivorous birds had statistically lower ($p \leq 0.05$) levels of selenium in liver and kidney but not in muscle ($p > 0.05$) than did birds that feed on fish and invertebrates. Grebes had statistically higher ($p \leq 0.05$) levels of selenium in muscle than any other species.

Table 6. Geometric means (GM) ± standard error (SE) and ranges of selenium concentrations (ppm, dry weight) in adult tissues by species across sites. Means with different capital letters are significantly different ($p \leq 0.05$) among tissues (n = number of birds analyzed).

Species	n	Liver GM±SE	Range	Kidney GM±SE	Range	Muscle GM±SE	Range
American coot	15	6.56 A ±1.10	3.4-12.8	12.13 A ±1.09	7.0-25.8	3.73 A ±1.12	1.6-7.7
Common moorhen	17	9.35 A ±1.12	4.1-16.2	12.29 A ±1.15	4.4-38.0	3.44 A ±1.19	1.2-7.3
Least bittern	27	14.92 B ±1.10	5.5-35.6	27.59 B ±1.12	8.3-63.2	3.73 A ±1.28	0.7-8.2
Green heron	10	15.99 B ±1.09	11.1-25.5	22.34 B ±1.12	13.5-48.5	4.6 A ±1.14	1.9-7.0
Pied-billed grebe	3	26.10 B ±1.13	22.5-28.5	25.52 B ±1.32	14.7-36.3	10.33 B ±1.27	8.1-13.2
F-Value		$F = 20.78$		$F = 8.13$		$F = 1.54$	

Age Comparisons:

Small sample sizes and the inability to collect all age classes across all species make generalizations concerning selenium as a function of life history stage difficult. As a result, no statistical tests were run, however, certain trends were apparent. Four moorhen chicks, one juvenile and two adults were collected from Bee Lake (backwater lake). Chicks had the highest levels of selenium in the liver followed by adults. The lowest levels occurred in the juvenile. One coot chick and five adults were collected from McAllister Lake (seep lake). The coot

chick had higher levels of selenium in the liver and kidney than the five adults. Nine juvenile birds were collected (four moorhens, three grebes, and two bitterns). Juvenile moorhens and grebes generally had lower selenium levels in liver, kidney, and muscle than adults of the same species collected at the same sites. In contrast, juvenile bitterns had higher liver selenium levels than adult bitterns but lower kidney and muscle levels. Overall, chicks had the highest selenium levels in the liver of any age class. Juveniles had the lowest liver concentrations in backwater lakes but these levels were higher than those in the livers of adults from seep lakes (Table 7).

Table 7. Geometric means (GM) of selenium concentrations (ppm, dry weight) ± standard error (SE) in livers of adults, juveniles, and chicks from backwater and seep lakes, 1993 (n = number of birds analyzed).

		Backwater Lakes		Seep Lakes	
Species	Age	n	GM ± SE	n	GM ± SE
Coot	Adult	6	9.2 ± 1.0	9	5.2 ± 1.1
	Chick	0	---	1	11.0
Moorhen	Adult	9	13.7 ± 1.0	8	6.1 ± 1.1
	Juvenile	4	7.6 ± 1.1	0	---
	Chick	4	15.5 ± 1.1	0	---
Bittern	Adult	13	21.9 ± 1.1	14	10.5 ± 1.1
	Juvenile	1	42.8	1	72.7
Heron	Adult	7	17.7 ± 1.1	3	12.6 ± 1.1
Grebe	Adult	3	26.1 ± 1.1	0	---
	Juvenile	3	11.7 ± 1.3	0	---

Eggs and Egg Masses:

Three completely formed eggs were collected. Two were lesser nighthawk (*Chordeiles acutipennis*) eggs from a nest near Butler lake (seep lake). The third was taken from the oviduct of an adult green heron. Selenium levels in the nighthawk eggs were 4.79 and 5.58 ppm, and the concentration in the heron egg was 8.68 ppm (Table 8). Developing egg masses (egg masses consisted of the ovary and the cluster of developing eggs surrounding the ovaries) were also collected from bitterns, grebes, and herons. The selenium concentrations in these egg masses ranged from 7.11 to 10.3 ppm. No developing eggs were found in coots or moorhens.

Table 8. Selenium concentrations (ppm, dry weight) in eggs and egg masses from several bird species and several sites on Imperial National Wildlife Refuge (NC=not collected).

Species	Collection Site	Egg	Egg Mass
Lesser nighthawk	Butler Lake	4.8	NC
Lesser nighthawk	Butler Lake	5.6	NC
Green heron	Adobe Lake	8.7	NC
Green heron	Adobe Lake	NC	10.3
Green heron	Butler Lake	NC	8.6
Pied-billed grebe	Island Lake	NC	9.9
Least bittern	McAllister Lake	NC	8.7

Tissue Selenium Ratios:

Kidney:liver ratios for all except eight adult birds collected on Imperial NWR were greater than 1; indicating "normal" dietary concentrations of selenium (Ohlendorf et al. 1990). Kidney and liver levels were not highly correlated in bitterns ($r=0.4918$, $p>0.05$) or moorhens ($r=0.2144$, $p>0.05$), however, they were correlated ($r=0.7491$, $p\leq 0.05$) in coots. Kidney and muscle correlations of selenium levels also varied with species. They were significantly correlated in coots ($r=0.7068$, $p\leq 0.05$) and bitterns ($r=0.6722$, $p\leq 0.05$), but not in moorhens ($r=0.2689$, $p>0.05$). Levels of selenium in liver and muscle were significantly correlated in bitterns ($r=0.8288$, $p\leq 0.05$), moorhens ($r=0.9137$, $p\leq 0.05$), and coots ($r=0.9179$, $p\leq 0.05$).

Dietary Comparisons:

Bitterns primarily fed on freshwater shrimp (*Paleomonetes* spp.) and crayfish (*Procambarus clarkii*). Herons preyed upon fish, i.e. threadfin shad (*Dorosoma petenense*), bluegill (*Lepomis macrochirus*) and mosquitofish (*Gambusia affinis*) (Table 9).

Table 9. Stomach contents of least bitterns and green herons across sites (n = number of birds in which prey were present in the gastrointestinal tract).

Species		Stomach Content				
	n	Shrimp	Crayfish	Threadfin shad	Bluegill	Mosquitofish
Least bittern	20	135	7	2	13	18
Green heron	10	6	6	22	14	11

Geometric means of selenium levels for fish and invertebrates commonly consumed as prey by herons and bitterns were reported by Lusk (1993) (Table 9). Lusk's (1993) samples were collected from the same sites as birds collected in this study. All prey organisms were above the 3 ppm minimum threshold for which toxic effects on predatory fish or birds might be expected.

Table 10. Geometric means of selenium concentrations (ppm, dry weight) of prey species collected by Lusk (1993) in 1991-1992, (NC=not collected).

	Seep Lakes		Backwater Lakes		
Prey Item	McAllister	Butler	Bee	Island	Adobe
Shrimp	7.2	3.3	5.9	8.9	10.9
Crayfish	4.2	2.4	20.8	10.3	17.1
Threadfin shad	NC	NC	4.2	5.9	4.4
Bluegill	NC	2.6	7.1	10.1	3.2
Mosquitofish	4.9	2.8	NC	10.4	NC

Exposure Time:

Exposure time was determined using the selenium concentrations in prey species reported by Lusk (1993) (Table 10). Assuming that the selenium concentrations in prey were constant between years (1992 and 1993), prey/tissue ratios were used to determine exposure time. The levels of selenium in the tissues indicated that some birds sampled had been acutely exposed, whereas others had been subjected to chronic exposures (Table 11). Eight of the nine bitterns (McAllister Lake) which fed primarily on shrimp (7.24 ppm selenium) had liver selenium levels greater than 7.24 ppm but had selenium concentrations in muscle less than 7.24 ppm. These results would indicate that the birds have been feeding at McAllister Lake more than seven days but less than 81 days (Heinz 1990). One bittern collected at McAllister Lake had both liver and muscle selenium concentrations less than 7.24 ppm. These levels would suggest that this bird had only recently begun feeding at this lake. Of the thirty-one birds for which selenium concentrations of prey species were known, seven had liver and muscle selenium levels greater than those in the prey species. These results would suggest that these birds had been feeding at these sites for more than 81 days. The remaining twenty-four birds had liver selenium levels greater than but muscle selenium levels less than those in prey species.

Table 11. The number of adult birds of two species for which exposure time was calculated based on selenium levels in liver and muscle.

Species	Acute Exposure	Chronic Exposure
Least bittern	22	3
Green heron	2	4

DISCUSSION

The paradox of selenium is that it is nutritionally essential in small amounts yet highly toxic in slightly higher amounts. For this reason, selenium has been difficult to study, especially in natural systems. The history of selenium in the lower Colorado River system is not well documented, however a portion of selenium input is naturally occurring.

Gender differences in selenium concentration have been noted in laboratory studies of mallards; males generally accumulated more selenium during the breeding season in livers than females (Heinz et al. 1987, 1989, 1990). Despite these reported differences under laboratory conditions, there are no data to indicate differences in selenium concentrations in liver based on gender during the breeding season under field conditions (Ohlendorf et al. 1990, Custer and Mitchell 1991, Burger and Gochfeld 1992). Results of this study are consistent with other field studies; there were no differences in liver selenium concentrations between sexes.

Species Trends:

The primary fall and winter dietary items of coots on the lower Colorado River consisted of algae (*Pithophora* sp.), sago pondweed (*Potamogeton pectinatus*) and cattail (*Typha latifolia*). Invertebrates were also taken during the breeding season (Eley and Harris 1976). Moorhens collected in Florida were primarily

herbivorous, but fed heavily on insects during the breeding season (Mulholland and Percival 1982). Stomach content of bitterns and herons collected during the present study suggested that bitterns primarily fed on freshwater shrimp and crayfish, whereas herons preyed upon fish, i.e., threadfin shad, bluegill and mosquitofish.

Linear contrasts indicated that selenium levels in liver and kidney were significantly lower in plant-eating species than in those feeding on animal matter. There was not a significant difference in selenium levels in muscle tissue between herbivores and carnivores. Since muscle accumulates selenium at a slower rate than other tissues, low levels could be indicative of short residence time (Heinz 1990).

All grebes collected in the present study had high selenium concentrations in their tissues. Eared grebes (*Podiceps nigricollis*) at Kesterson NWR also had high levels of selenium in their tissues (Ohlendorf et al. 1986a). Grebes are divers that feed primarily on aquatic invertebrates and fish (Ehrlich et al. 1988). They may also feed on aquatic invertebrates from the bottom of the lake. Detritus is abundant at the sediment-water interface and often contains high concentrations of selenium (Lusk 1993). Ingesting detritus during feeding could expose grebes to very high concentrations of selenium.

Herons also had high levels of selenium in liver and muscle. Selenium levels in liver and muscle of least bitterns were very close to those in herons.

Similarity is probably due to the common diet (Ehrlich et al. 1988). Despite these similarities, kidney concentrations were higher in bitterns than in herons. The reasons for these differences are unclear. However, the small mass of the bittern kidneys caused detection limits to be higher in bitterns than in herons. Therefore, differences in laboratory analysis may be a causal factor.

Selenium levels were similar in kidney and muscle for both herbivorous species. However, moorhens had higher concentrations of selenium in livers than did coots. Differences may be related to diet. Ohlendorf et al. (1990) indicated that there were no consistent patterns in selenium levels between species due to food habits at Kesterson NWR. Dubowy (1989) suggests that although herbivorous species feed on foods low in selenium, their daily caloric intake and hence, mass and amount of selenium is much higher than those that feed on aquatic invertebrates and fish. Despite these assertions, the data from the lower Colorado River consistently show lower selenium levels in herbivores than carnivores (Lusk 1993, Welsh 1992, data from the current study). If selenium levels in plants are significantly lower than in animal food items, then the proportion of vegetation that would have to be consumed in order to attain similar selenium levels in tissues would be very large. Lusk (1993) hypothesized a detrital food chain mechanism of selenium uptake because detritus contained higher levels of selenium than plants. If this hypothesis were true, birds feeding on detritus or detritivores would have a higher intake of selenium than those

feeding on plants. The data from the current study tend to support this hypothesis.

Although Kesterson has been the model for point source selenium contamination, the cycling of selenium may be different in a non-point source system such as the lower Colorado River. Unlike Kesterson NWR which had selenium concentrations in water of 300 ppb, the lower Colorado River water contains only 2 ppb. Selenium contamination at Kesterson was from sub-surface irrigation drainwater of seleniferous soils. Selenium in the Colorado River comes from many sources, some naturally occurring, in the upper basin. Therefore, amount of selenium accumulated in tissues versus dietary selenium levels, and accumulation and elimination rates, may vary between species at Kesterson NWR and the lower Colorado River.

Habitat Differences:

Colorado River water enters seep lakes only after being filtered through the soil column. Filtration may remove organic selenium compounds. Conversely, backwater lakes receive water via a direct connection to the river. Therefore, selenium levels are constantly being augmented in backwater lakes. All species of aquatic birds collected on backwater lakes had higher selenium levels in the three tissues sampled than those collected on seep lakes. Birds collected on backwater lakes had liver concentrations that were twice those of

birds collected on seep lakes. In addition, muscle selenium concentrations for birds collected on backwater lakes were almost three times the levels of those collected on seep lakes. During the breeding season, birds tend to conduct their daily activities close to the nest (Piest and Sowls 1985). Adults feeding young tend to capture prey as close to the nest as possible (Ehrlich et al. 1988) to maintain energetic efficiency. It would be more efficient for adults to feed young birds food taken from the same body of water in which they build their nests. In this study there was a significant difference in tissue selenium levels between birds collected on seep lakes and birds collected on backwater lakes, suggesting that birds had been feeding at the site in which they were collected. Therefore, birds appeared to have significant fidelity to feeding sites. Maintenance of energetic efficiency while rearing young could explain these data.

Species-Habitat Trends:

Of all aquatic birds collected, fish- and invertebrate-eating species from backwater lakes had the highest selenium concentrations in tissues. Carnivorous birds had higher tissue selenium levels than did those that feed on vegetation in both seep lakes and backwater lakes. However, selenium levels in both fish- and vegetation-feeders collected from backwater lakes were higher than those collected from seep lakes. Birds feeding on plants in backwater lakes had comparable or higher selenium levels in liver and muscle than did fish-eating

birds collected from seep lakes. Despite the fact that selenium levels in liver were lower in birds from seep lakes than those from backwater lakes, levels in some fish-eaters were still at or above the 10 ppm threshold level indicating that reproductive success is at risk because of selenium-induced teratogenesis (Lemly 1993). Selenium levels in vegetative feeders from seep lakes were all below the 10 ppm concern level. These data suggest that there is a difference in the risk of selenium exposure based on feeding site as well as diet.

Age Differences:

Although chicks collected from backwater lakes had higher selenium levels in their tissues than those from seep lakes, young birds seem to have a propensity to concentrate selenium in their tissues at all sites. A coot chick collected at McAllister Lake had one of the highest selenium levels in liver and kidney of any of the coots collected. The four moorhen chicks collected on Bee Lake had liver selenium levels similar to those in the two adult moorhens collected at the same site. In contrast, the juvenile moorhen collected at Bee Lake had the lowest level of liver selenium of any age class taken from that site. Chicks are flightless and their foraging range is fairly small, therefore it appears reasonable that tissue selenium concentrations should reflect site-specific environmental selenium levels.

Generally, juveniles had lower selenium levels than chicks or adults collected at the same site. Juvenile grebes, coots, and moorhens had lower tissue

selenium levels than adults collected at the same sites but juvenile bitterns had higher levels of selenium in the liver than adults. The trend of higher selenium levels in the tissues of birds taken from backwater lakes than those taken from seep lakes also occurred in juveniles; juvenile moorhens reared on backwater lakes had higher selenium levels than adults collected on seep lakes.

Egg and Egg Mass Trends:

Lemly (1993) suggests that measuring selenium concentrations in gravid ovaries and eggs is the most precise way to evaluate potential reproductive impacts to aquatic bird populations. Selenium concentrations in gravid ovaries follow the same trend as the concentrations in adult tissues; fish-eaters from backwater lakes had higher selenium levels in gravid ovaries than did fish-eaters from seep lakes. However, the selenium levels in gravid ovaries from both habitats were above the 3 ppm effect threshold. This effect threshold was proposed by Lemly (1993) as the level at which selenium impacts on avian reproduction would likely occur. In addition, the selenium concentration in the single heron egg taken, was well above the 3 ppm effect threshold. This result raises the possibility of teratogenesis in the offspring of fish-eating birds nesting on backwater lakes.

Selenium levels in eggs of lesser nighthawks were also above the 3 ppm effect threshold for reproductive impairment. These results suggest that

bioaccumulation of selenium may also occur in neotropical migrants nesting on Imperial NWR. Lesser nighthawks are insectivorous and feed on emerging aquatic insects. These insects are known to have elevated selenium level in their aquatic phase (Lusk 1993) and would appear to carry these levels with them into the aerial phase. Therefore, neotropical migrants feeding on emerging aquatic insects might be exposed to elevated selenium levels. Since many birds feed in the riparian area, bioaccumulation of selenium through the aquatic food chain would result in high selenium concentrations in eggs of several species of neotropical migrants. The bioaccumulation of selenium in tissues and eggs of insectivorous birds nesting near selenium-contaminated habitats have previously been documented (King et al. 1994, Ohlendorf et al. 1990).

Kidney:Liver Ratios:

Tissue ratios of selenium concentrations have been used as a quick method of assessing selenium exposure histories. Seventy of eighty-one birds in this study had kidney:liver ratios above one. It has been assumed that selenium content in kidneys exceeds that in livers when dietary levels are low (Goede 1985). These ratios would suggest dietary levels were "normal" based on the criteria of Ohlendorf et al. (1990). However, selenium levels for food chain organisms of the aquatic bird species collected for this study were above the effect threshold for potential lethal effects on consumers (Lusk 1993). In addition, liver selenium

concentrations were above the effect threshold for adverse biological effects in aquatic birds, and were above the threshold for teratogenesis in eggs (Skorupa et al. 1992, Lemly 1993). Despite the levels in the tissues and the eggs, kidney:liver ratios indicated that there is no cause for concern. Kidney and liver selenium levels were not highly correlated in bitterns or coots. Lack of correlation suggests that kidney:liver ratios would not give a reliable indication of risk. Therefore, it appears that kidney:liver selenium ratios are not a reliable indicator of risk to birds along the lower Colorado River and absolute tissue levels should be considered. Results from this study would also suggest using these ratios with caution.

Exposure Time:

Using tissue accumulation and elimination rates from Heinz (1990), and selenium levels in prey items from Lusk (1993), the majority of birds collected had acute rather than chronic exposure to elevated selenium levels. Acute exposure to selenium suggests that the birds had been feeding at a particular site for more than seven days but less than 81 days. The results of the liver:muscle comparison indicate that a bird feeding at a particular site for more than 81 days had chronic exposure. However, there are several limitations in this analysis as conducted. Information determined under laboratory conditions do not always reflect what occurs under field conditions; e.g. gender differences in selenium

concentrations in field and laboratory settings. In addition, selenium levels in prey items were estimated from data collected at the same sites but a year earlier than the current study. There are no data to suggest that selenium exposure levels change over time, but it is possible that they do. Therefore, the hypotheses concerning exposure history and the consistency of selenium levels over time must be further evaluated before definitive statements can be made.

CONCLUSIONS

Tissue levels in birds collected from the lower Colorado River are above the biological toxic effect thresholds for the health and reproduction of aquatic birds. Selenium levels in tissues of aquatic birds on Imperial NWR seem to indicate acute exposure to elevated selenium levels. There were differences between age classes in tissue selenium levels. Chicks had higher selenium levels in liver than adults of the same species collected on the same site. There were also differences in tissue selenium concentrations based on diet. Those species that feed primarily on animal matter had higher selenium concentrations in their tissues than those that feed on aquatic plants.

There were no temporal differences in the selenium levels in the tissues of any species; levels in individuals collected at the beginning of the breeding season were comparable to those collected at the end of the breeding season.

There were no significant gender differences in tissue selenium levels but there were significant differences between habitat types. Birds taken from backwater lakes had significantly higher selenium levels in their tissues than those taken from seep lakes, regardless of species. Tissue levels of selenium were not at levels of concern in herbivorous species feeding on seep lakes but they were at levels of concern for birds that primarily feed on fish and invertebrates from seep lakes and for all bird species collected from backwater lakes. Further study is needed to understand the mechanism by which certain avian species accumulate

such high levels of selenium and what effects these levels have on reproductive success.

MANAGEMENT IMPLICATIONS

High selenium levels in the lower Colorado River valley have significant implications to resident and breeding populations of birds. The following are recommendations and suggestions:

▲ Seep lakes currently offer habitats with significantly lower levels of selenium than those present in backwater lakes. Connecting seep lakes to the river may result in elevated selenium levels similar to those in backwater lakes.

▲ Tissue selenium levels approach or are above the level of concern, but there are no data to document the effects of high selenium levels on waterbird reproductive success or teratogenesis under the conditions peculiar to the lower Colorado River. High priority should be given to studies that document the effects of existing selenium levels on reproductive success and teratogenesis.

▲ Fish- and invertebrate-eating birds had the highest levels of selenium in their tissues. Further studies should investigate the effects of food habits on selenium exposure given the conflicting conclusions of Ohlendorf et al. (1988). One suggestion would be to release pinioned birds on backwater lakes and seep lakes and evaluate reproductive success and teratogenesis.

- It is highly likely that insects newly emerged from sediments with elevated selenium levels are carrying potentially harmful concentrations of selenium. The data from nighthawk eggs may indicate that neotropical migrants that feed on insects emerging from the backwater lakes are also being exposed to elevated selenium levels. Further studies are needed to define potential effects of the bioaccumulation of selenium in bats and non-aquatic avian species feeding along the lower Colorado River.

APPENDIX A

Analytical Results

Table A-1. Analytical results (μg/g, dry weight) in tissues of all birds collected at McAllister Lake, along with sex, age, and whole body weight of each bird, date of collection (1993), and percent (%) moisture. (NA = sex unknown or not enough tissue for chemical analysis.)

Common Name	Sex	Age	Whole Body Weight (g)	Date	% Moisture Liver	Liver	% Moisture Kidney	Kidney	% Moisture Muscle	Muscle
American coot	NA	Chick	107.3	5-25	73.2	11.00	78.0	15.86	NA	NA
American coot	♂	Adult	600.0	5-25	74.8	9.03	78.1	11.05	75.9	7.47
American coot	♂	Adult	420.0	5-25	70.4	5.01	84.3	15.36	76.4	2.47
American coot	♀	Adult	440.0	5-26	70.9	5.9	76.4	8.76	74.3	3.31
American coot	♂	Adult	500.0	5-27	72.0	4.27	74.2	11.04	74.8	2.93
American coot	♂	Adult	610.0	5-27	67.6	6.67	75.8	11.48	69.3	3.88
Common moorhen	♀	Adult	248.6	5-26	69.5	6.05	90.6	25.52	75.3	1.61
Common moorhen	♀	Adult	260.0	5-27	71.3	7.19	80.8	12.3	74.5	2.53
Common moorhen	♀	Adult	280.0	5-27	69.3	8.96	85.7	9.15	76.7	1.87
Least bittern	♀	Adult	85.4	4-30	69.1	8.41	NA	NA	71.5	1.09
Least bittern	♀	Adult	97.8	4-30	68.5	16.30	91.4	30.59	69.7	4.03
Least bittern	♀	Adult	85.0	4-30	71.8	11.80	91.2	24.63	71.3	3.37
Least bittern	♀	Adult	91.9	5-01	68.1	14.3	97.1	62.38	71.5	4.73
Least bittern	♀	Adult	106.4	5-25	67.7	13.3	83.7	63.21	74.1	3.8
Least bittern	♂	Adult	77.9	5-25	69.1	20.1	NA	NA	73.7	2.12
Least bittern	♀	Adult	100.1	5-25	72.0	6.92	92.9	9.787	73.1	0.69
Least bittern	♀	Adult	97.5	5-25	72.7	9.98	49.9	16.72	71.5	2.61
Least bittern	♀	Adult	108.9	5-26	68.6	10.2	86.1	8.33	67.0	1.84

Table A-2. Analytical results (μg/g, dry weight) in tissues of all birds collected at Butler Lake, along with sex, age, and whole body weight of each bird, date of collection (1993), and percent (%) moisture. (NA = sex unknown or not enough tissue for chemical analysis.)

Common Name	Sex	Age	Whole Body Weight (g)	Date	% Moisture Liver	Liver	% Moisture Kidney	Kidney	% Moisture Muscle	Muscle
American coot	♀	Adult	440.0	5-25	70.5	3.36	77.6	7.02	71.3	1.63
American coot	♀	Adult	700.0	5-25	74.0	5.22	83.2	NA	71.2	2.54
American coot	♂	Adult	750.0	5-27	71.4	4.49	80.5	9.78	68.9	2.51
American coot	♂	Adult	610.0	5-27	71.4	5.06	79.4	8.3	71.4	2.7
Common moorhen	♂	Adult	370.0	5-25	70.6	6.44	72.4	14.1	74.2	2.14
Common moorhen	♂	Adult	360.0	5-27	72.7	5.08	77.2	6.07	72.3	1.65
Common moorhen	♂	Adult	385.0	5-27	68.7	4.07	72.9	8.74	73.2	1.26
Common moorhen	♀	Adult	300.0	5-27	74.2	7.07	71.9	9.05	74.9	1.19
Common moorhen	♂	Adult	325.0	5-27	72.7	4.98	67.6	9.6	74.0	NA
Least bittern	♀	Adult	113.4	5-25	73.9	8.12	35.0	13.16	72.4	1.47
Least bittern	♀	Adult	78.6	5-27	70.8	7.18	81.9	23.00	72.6	1.64
Least bittern	♀	Adult	90.4	5-27	70.6	18.00	59.2	15.00	71.9	4.68
Least bittern	♀	Adult	77.0	5-27	70.2	5.53	83.9	21.11	73.1	1.21
Least bittern	♂	Adult	102.6	5-27	70.6	7.35	43.4	19.35	73.4	2.55
Least bittern	♂	Juvenile	74.6	5-28	73.6	72.70	60.7	6.14	72.6	1.59
Green heron	♂	Adult	219.0	5-02	71.6	15.09	67.7	22.30	70.4	1.88
Green heron	♂	Adult	245.8	5-27	92.0	11.96	64.6	13.53	74.7	3.86
Green heron	♀	Adult	197.0	5-27	69.2	11.14	65.9	19.28	72.7	3.38

Table A-3. Analytical results (μg/g, dry weight) in tissues of all birds collected at Island Lake, along with sex, age, and whole body weight of each bird, date of collection (1993), and percent (%) moisture. (NA = sex unknown or not enough tissue for chemical analysis.)

Common Name	Sex	Age	Whole Body Weight (g)	Date	% Moisture Liver	Liver	% Moisture Kidney	Kidney	% Moisture Muscle	Muscle
American coot	♂	Adult	450.0	6-02	69.5	9.10	55.0	11.79	71.7	3.62
American coot	♀	Adult	415.7	6-10	71.5	7.25	79.6	15.02	88.4	4.98
American coot	♂	Adult	425.0	7-23	75.8	8.80	76.8	14.14	74.4	5.3
American coot	♂	Adult	623.0	7-23	77.0	8.38	76.8	14.83	74.4	5.54
Common moorhen	♂	Adult	320.0	6-02	67.0	14.7	75.7	4.43	76.4	6.67
Common moorhen	♂	Adult	328.8	6-03	67.1	12.83	76.9	12.6	72.2	3.75
Common moorhen	♀	Adult	332.5	6-03	70.6	16.19	75.4	5.46	71.9	5.41
Common moorhen	♂	Adult	361.1	6-23	69.3	13.95	69.5	38.03	74.2	6.3
Common moorhen	♀	Juvenile	219.8	7-23	69.3	6.25	80.9	12.99	72.5	3.05
Least bittern	♂	Adult	98.4	6-03	68.2	22.7	90.5	54.18	72.2	6.28
Least bittern	♀	Adult	98.6	6-03	70.0	17.9	91.1	60.86	70.9	4.8
Least bittern	♀	Adult	97.1	6-03	68.0	34.2	83.2	23.82	72.0	5.76
Least bittern	♀	Adult	86.3	6-04	67.6	21.00	76.2	31.56	72.7	7.7
Least bittern	♂	Adult	83.3	6-04	69.0	23.5	73.3	24.84	70.9	7.46
Least bittern	♀	Adult	103.8	6-07	71.0	23.9	80.0	28.57	71.3	6.61
Least bittern	NA	Juvenile	54.3	6-07	74.4	42.8	NA	NA	NA	NA
Least bittern	♂	Adult	77.3	6-07	71.1	21.6	87.8	38.6	69.3	4.75
Green heron	♀	Adult	230.1	6-07	70.0	21.4	73.2	18.8	73.4	6.37
Pied-billed grebe	♀	Adult	286.9	6-07	74.4	22.5	75.0	14.67	69.1	8.1
Pied-billed grebe	♀	Adult	345.0	6-07	69.9	27.6	NA	98.03	74.0	9.88
Pied-billed grebe	♀	Juvenile	233.0	6-23	69.9	6.83	86.0	25.43	71.0	2.2

Table A-4. Analytical results ($\mu g/g$, dry weight) in tissues of all birds collected at Bee Lake, along with sex, age, and whole body weight of each bird, date of collection (1993), and percent (%) moisture. (NA = sex unknown or not enough tissue for chemical analysis.)

Common Name	Sex	Age	Whole Body Weight (g)	Date	% Moisture Liver	Liver	% Moisture Kidney	Kidney	% Moisture Muscle	Breast Muscle
Common moorhen	♀	Adult	263.2	6-04	67.0	14.29	70.5	17.2	74.0	6.59
Common moorhen	♂	Adult	319.9	6-04	67.8	15.58	60.3	21.4	73.1	6.77
Common moorhen	NA	Chick	31.5	6-04	50.0	13.16	NA	NA	NA	NA
Common moorhen	NA	Chick	41.8	6-04	64.2	15.24	83.7	63.5	NA	NA
Common moorhen	NA	Chick	50.2	6-10	70.5	19.26	NA	NA	NA	NA
Common moorhen	NA	Chick	46.2	6-10	72.1	14.75	NA	NA	NA	NA
Common moorhen	♂	Juvenile	257.0	8-06	70.7	10.00	67.7	16.2	77.6	5.54
Least bittern	♂	Adult	91.6	6-10	71.8	23.57	85.5	NA	73.7	5.52
Least bittern	♂	Adult	80.4	6-16	63.9	22.41	73.7	NA	71.8	6.11
Least bittern	♂	Adult	77.1	7-22	70.5	20.69	72.5	31.9	71.9	6.37
Least bittern	♀	Adult	88.9	8-06	70.5	35.60	63.2	28.3	71.7	5.31
Green heron	♀	Adult	238.4	6-16	70.3	14.99	59.5	15.8	72.1	3.77
Green heron	♂	Adult	229.0	6-23	68.6	25.5	56.0	25.9	72.8	7.0
Green heron	♀	Adult	254.7	7-02	69.2	21.24	64.9	20.5	74.1	6.0
Pied-billed grebe	♀	Juvenile	323.6	6-10	71.6	14.45	69.7	21.6	71.8	<4.31

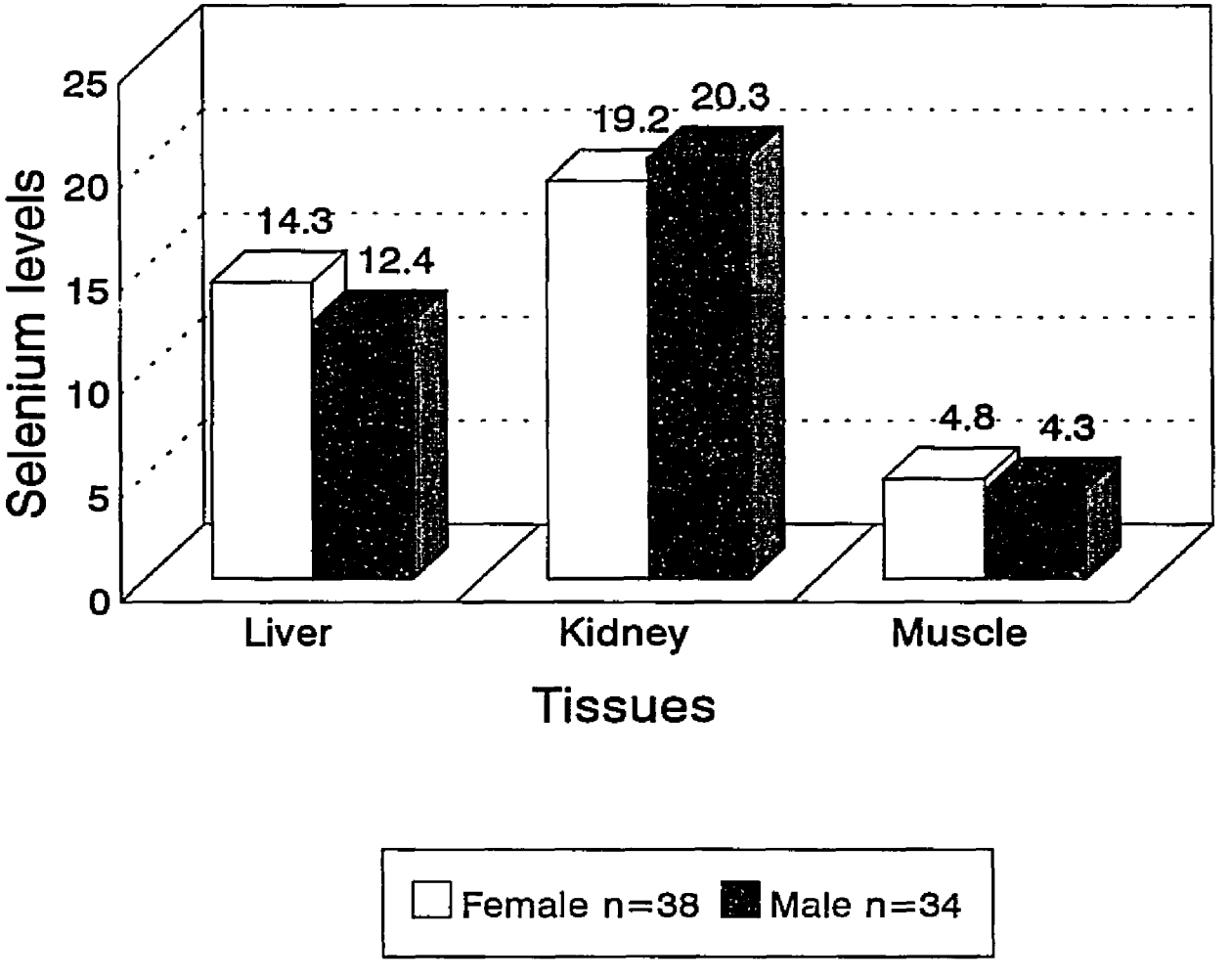
Table A-5. Analytical results (μg/g, dry weight) in tissues of all birds collected at Adobe Lake, along with sex, age, and whole body weight of each bird, date of collection (1993), and percent (%) moisture.

Common Name	Sex	Age	Whole Body Weight (g)	Date	% Moisture Liver	Liver	% Moisture Kidney	Kidney	% Moisture Muscle	Breast Muscle
American coot	♀	Adult	383.7	6-28	70.4	12.82	67.3	25.8	70.1	7.67
American coot	♂	Adult	471.0	8-04	71.9	9.41	76.4	14.6	74.2	4.78
Common moorhen	♂	Adult	381.6	6-28	72.3	13.46	69.3	19.0	75.8	7.26
Common moorhen	♂	Adult	305.5	6-28	70.3	13.64	69.4	19.7	75.2	7.17
Common moorhen	♂	Adult	341.3	7-23	72.1	9.97	74.6	11.2	75.2	4.8
Common moorhen	♀	Juvenile	320.3	8-02	70.0	6.88	62.9	8.15	71.5	2.75
Common moorhen	♂	Juvenile	381.0	8-04	72.6	7.88	73.9	11.4	75.0	4.95
Least bittern	♂	Adult	99.7	6-11	71.49	14.84	56.9	27.84	70.39	3.87
Least bittern	♀	Adult	103.1	6-28	70.83	16.9	77.78	40.6	72.4	6.3
Least bittern	♀	Adult	86.3	7-01	67.8	28.5	76.79	53.35	75.39	8.23
Least bittern	♂	Adult	91.1	8-02	72.76	14.1	79.07	37.91	75.08	7.11
Green heron	♀	Adult	223.9	6-07	74.61	12.9	77.53	28.84	76.35	4.37
Green heron	♂	Adult	222.4	8-02	76.24	19.1	77.63	24.11	73.8	5.37
Green heron	♂	Adult	253.0	8-04	66.22	12.7	76.67	48.48	73.32	6.89
Pied-billed grebe	♀	Juvenile	265.6	7-29	73.52	16.2	77.37	23.0	32.95	11.94
Pied-billed grebe	♀	Adult	357.3	8-06	70.38	28.5	22.44	36.3	73.95	13.18

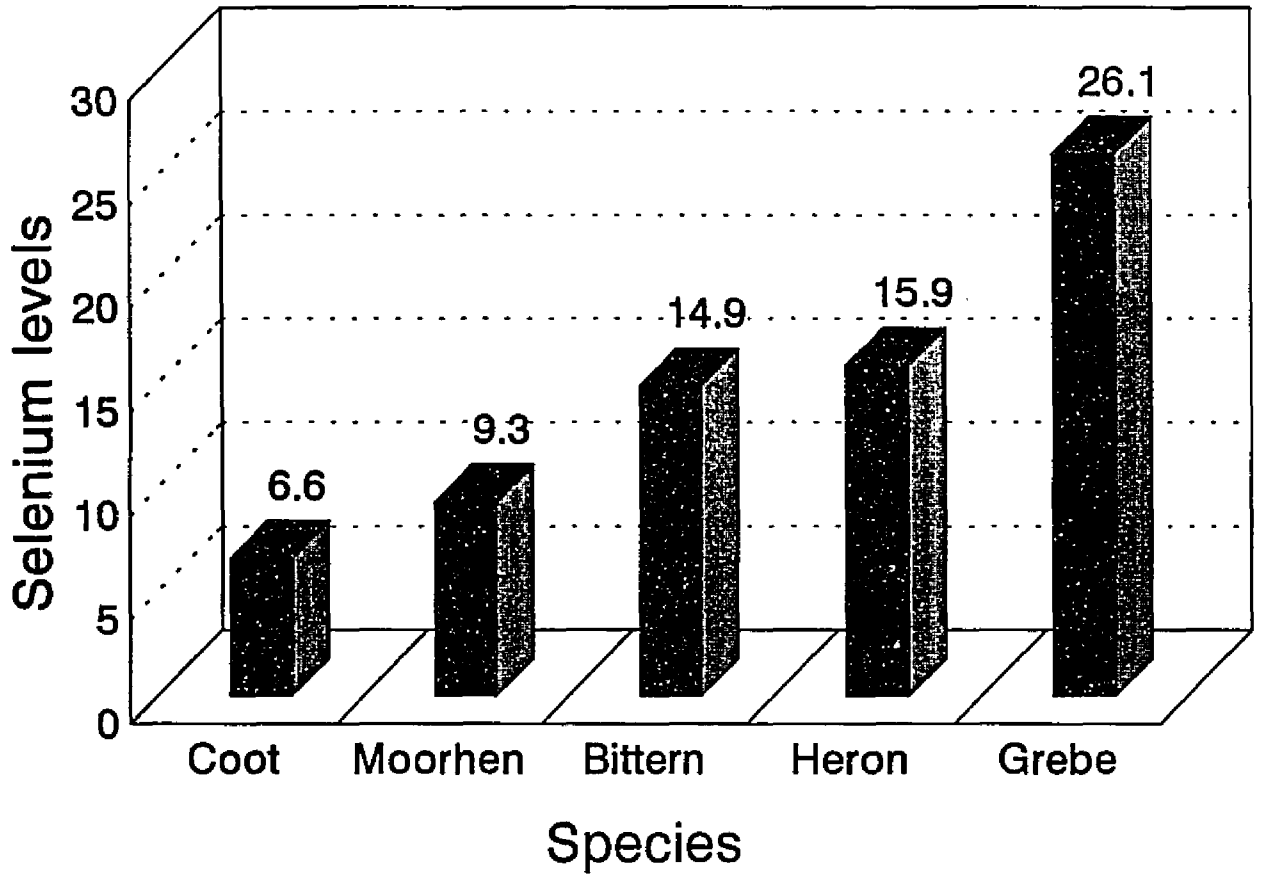
APPENDIX B

Graphs

B-1. Geometric means of selenium concentrations (μg/g, dry weight) in adult tissues by gender.

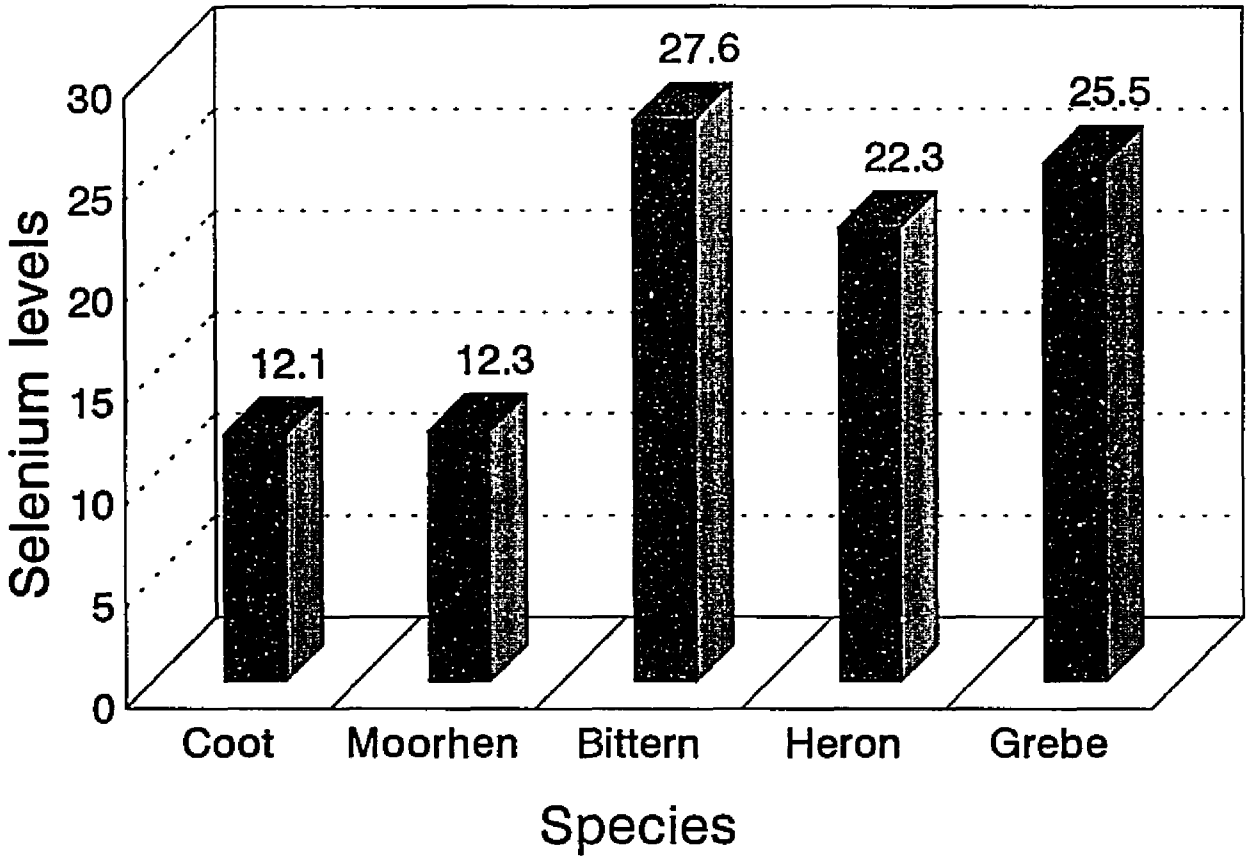


B-2. Geometric means of selenium concentrations (µg/g, dry weight) in adult livers by species across sites.



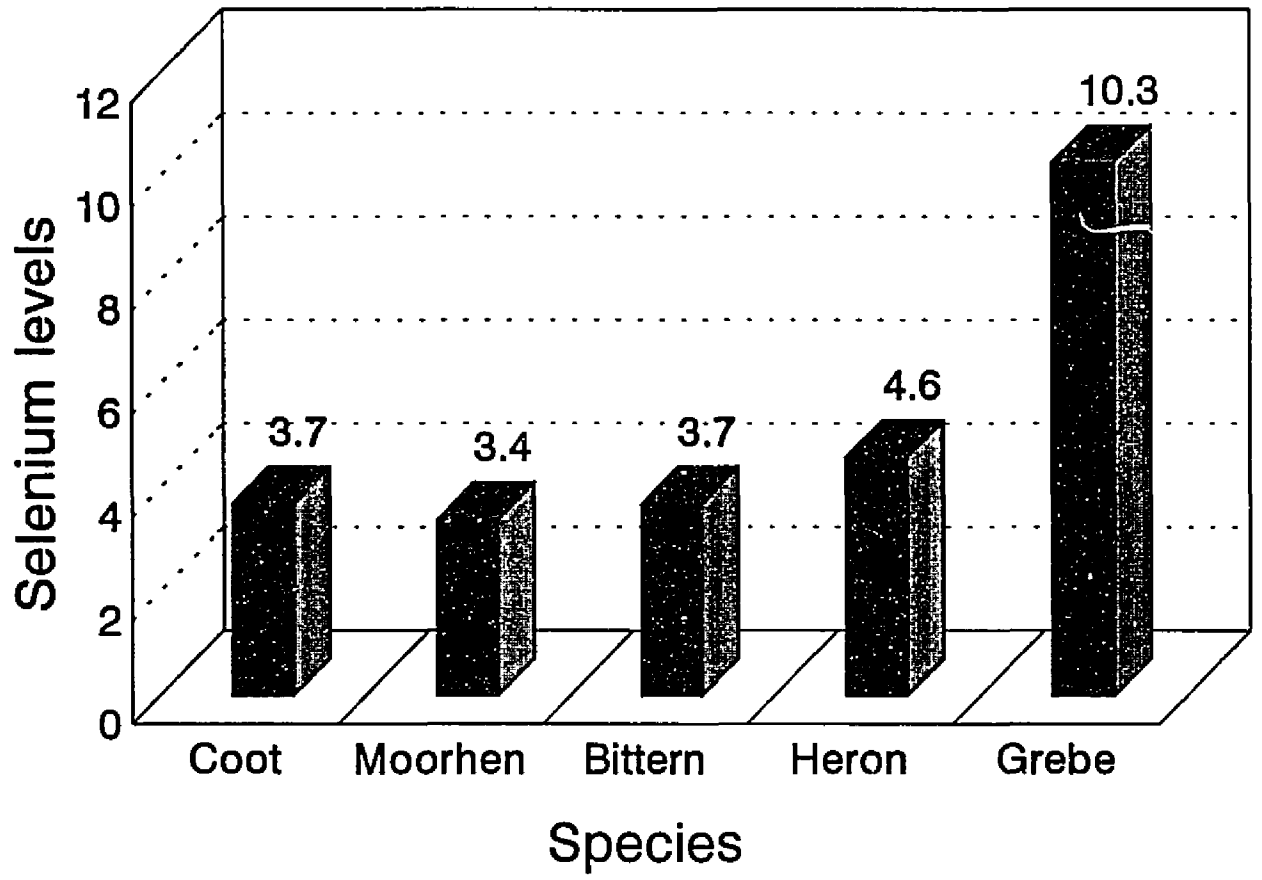
Coot n=15 Moorhen n=17 Bittern n=27 Heron n=10 Grebe n=3

B-3. Geometric means of selenium concentrations (μg/g, dry weight) in adult kidneys by species across sites.



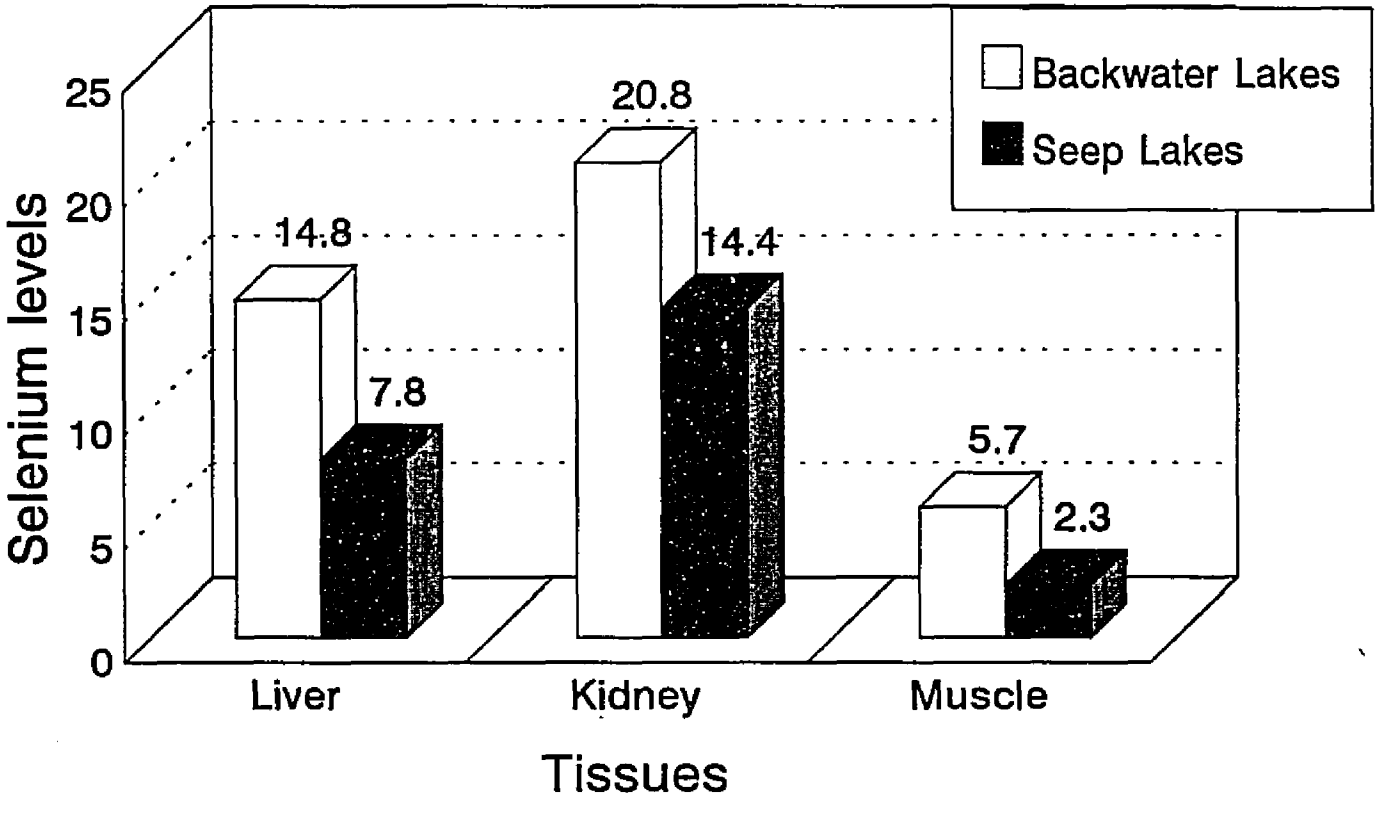
Coot n=15 Moorhen n=17 Bittern n=27 Heron n=10 Grebe n=3

B-4. Geometric means of selenium concentrations (μg/g, dry weight) in adult muscle by species across sites.



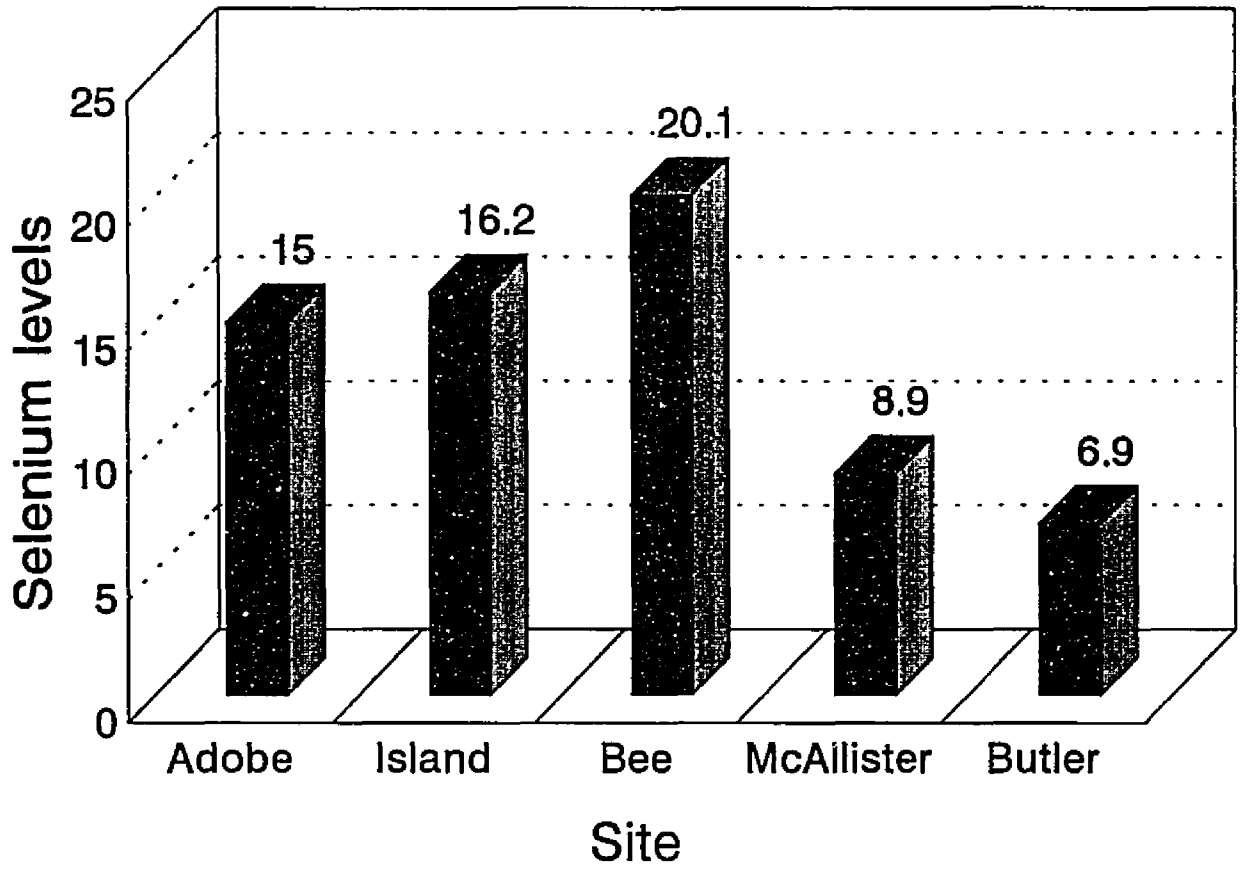
Coot n=15 Moorhen n=17 Bittern n=27 Heron n=10 Grebe n=3

B-5. Geometric means of selenium concentrations (μg/g, dry weight) of adult tissues for all species collected at each site class.



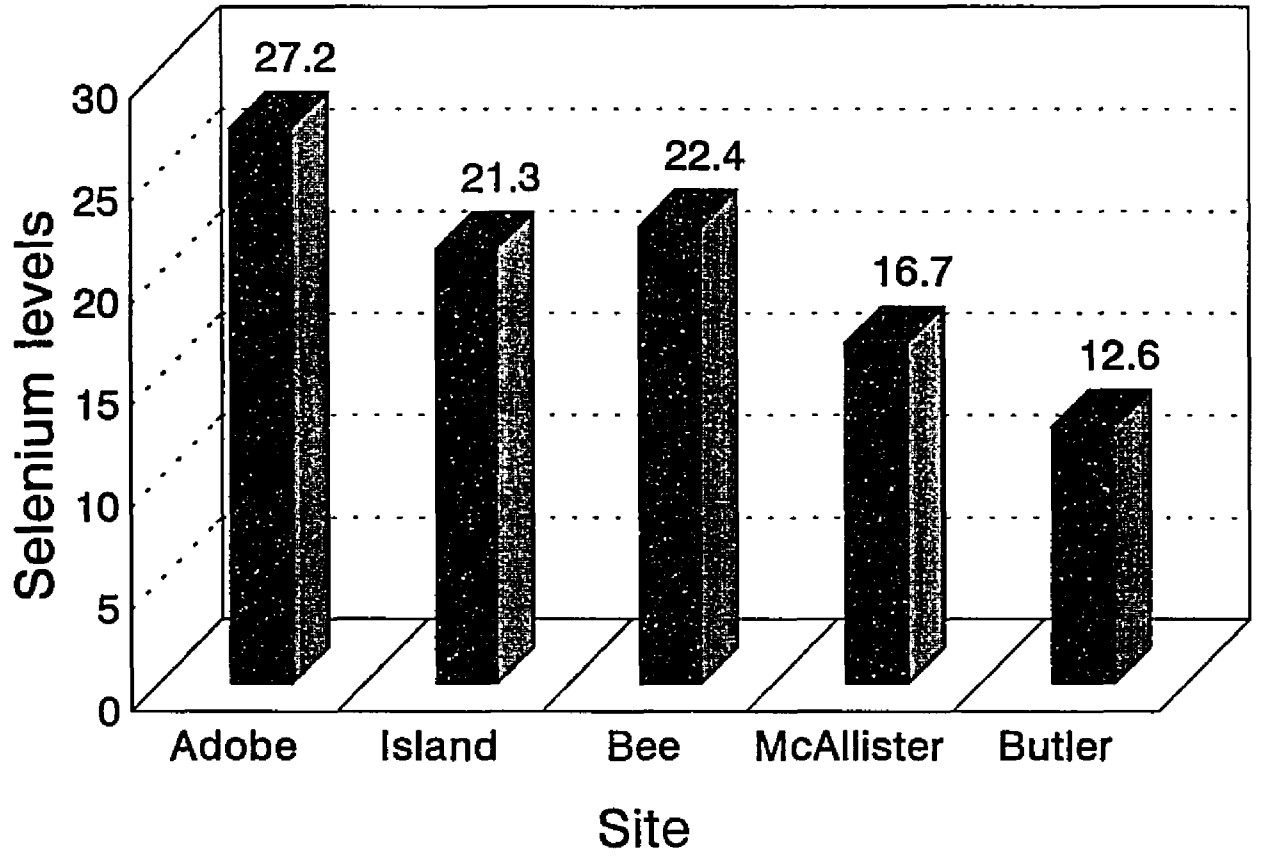
Birds collected on Backwater Lakes n=38
Birds collected on Seep Lakes n=34

B-6. Geometric means of selenium concentrations (μg/g, dry weight) in adult livers for all species by site.



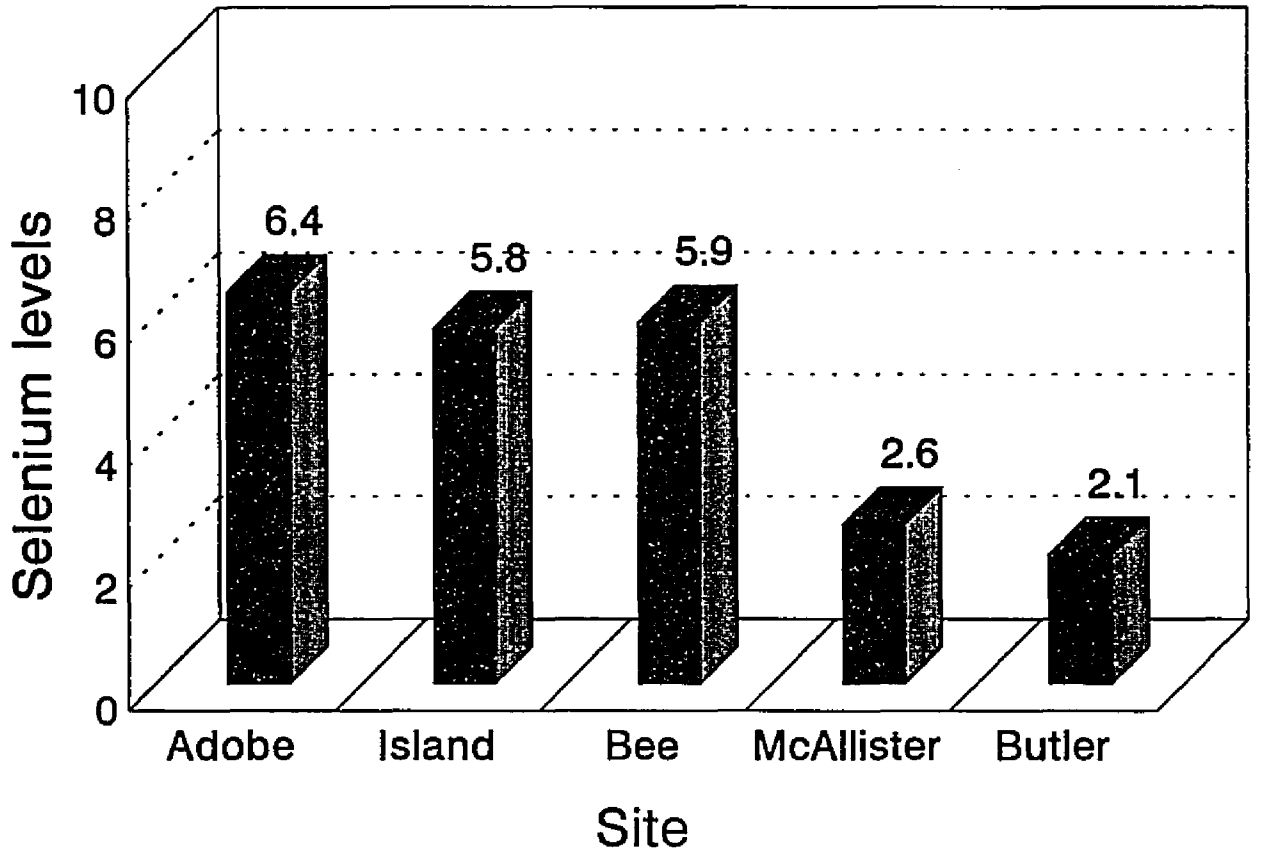
Adobe n=13 Island n=18 Bee n=7 McAllister n=17 Butler n=17

B-7. Geometric means of selenium concentrations (μg/g, dry weight) in adult kidneys for all species by site.



Adobe n=13 Island n=18 Bee n=7 McAllister n=17 Butler n=17

B-8. Geometric means of selenium concentrations (μg/g, dry weight) in adult muscle for all species by site.



Adobe n=13 Island n=18 Bee n=7 McAllister n=17 Butler n=17

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