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Environmental contaminants and biomarker responses in fish from the Columbia River and its tributaries: Spatial and temporal trends

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Abstract

Fish were collected from 16 sites on rivers in the Columbia River Basin (CRB) from September 1997 to April 1998 to document temporal and spatial trends in the concentrations of accumulative contaminants and to assess contaminant effects on the fish. Sites were located on the mainstem of the Columbia River and on the Snake, Willamette, Yakima, Salmon, and Flathead Rivers. Common carp (*Cyprinus carpio*), black bass (*Micropterus* sp.), and largescale sucker (*Catostomus macrocheilus*) were the targeted species. Fish were field-examined for external and internal lesions, selected organs were weighed to compute somatic indices, and tissue and fluid samples were preserved for fish health and reproductive biomarker analyses. Composite samples of whole fish, grouped by species and gender, from each site were analyzed for organochlorine and elemental contaminants using instrumental methods and for 2,3,7,8-tetrachloro dibenzo-*p*-dioxin-like activity (TCDD-EQ) using the H4IIE rat hepatoma cell bioassay. Overall, pesticide concentrations were greatest in fish from lower CRB sites and elemental concentrations were greatest in fish from upper CRB sites. These patterns reflected land uses. Lead (Pb) concentrations in fish from the Columbia River at Northport and Grand Coulee, Washington (WA) exceeded fish and wildlife toxicity thresholds (>0.4 μg/g). Selenium (Se) concentrations in fish from the Salmon River at Riggins, Idaho (ID), the Columbia River at Vernita Bridge, WA, and the Yakima River at Granger, WA exceeded toxicity thresholds for piscivorous wildlife (>0.6 μg/g). Mercury (Hg) concentrations in fish were elevated throughout the basin but were greatest (>0.4 μg/g) in predatory fish from the Salmon River at Riggins, ID, the Yakima River at Granger, WA, and the Columbia River at Warrendale, Oregon (OR). Residues of *p,p′*-DDE were greatest (>0.8 μg/g) in fish from agricultural areas of the Snake, Yakima, and Columbia River basins but were not detected in upper CRB fish. Other organochlorine pesticides did not exceed toxicity thresholds in fish or were detected infrequently. Total polychlorinated biphenyls (PCBs; >0.11 μg/g) and TCDD-EQs (>5 pg/g) exceeded wildlife guidelines in fish from the middle and lower CRB, and ethoxyresorufin *O*-deethylase (EROD) activity was also elevated at many of the same sites. Temporal trend analysis indicated decreasing or stable concentrations of Pb, Se, Hg, *p,p′*-DDE, and PCBs at most sites where historical data were available. Altered biomarkers were noted in fish throughout the CRB. Fish from some stations had responded to chronic contaminant exposure as

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indicated by fish health and reproductive biomarker results. Although most fish from some sites had grossly visible external or internal lesions, histopathological analysis determined these to be inflammatory responses associated with helminth or myxosporidian parasites. Many largescale sucker from the Columbia River at Northport and Grand Coulee, WA had external lesions and enlarged spleens, which were likely associated with infections. Intersex male smallmouth bass (*Micropterus dolomieu*) were found in the Snake River at Lewiston, ID and the Columbia River at Warrendale, OR. Male bass, carp, and largescale sucker containing low concentrations of vitellogenin were common in the CRB, and comparatively high concentrations (>0.3 mg/mL) were measured in male fish from the Flathead River at Creston, Montana, the Snake River at Ice Harbor Dam, WA, and the Columbia River at Vernita Bridge, WA and Warrendale, OR. Results from our study and other investigations indicate that continued monitoring in the CRB is warranted to identify consistently degraded sites and those with emerging problems.
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Keywords: Selenium; Mercury; Lead; Pesticides; Organochlorine chemicals; Ethoxyresorufin *O*-deethylase (EROD) activity; Health assessment index (HAI); Biomarkers; Ovotestis; Vitellogenin

1. Introduction

The Columbia River (CR) is the largest river in the Pacific Northwest and the fourth largest river in the US. The CR flows 1955 km and drains approximately 670,800 km^2. The CR has an average annual discharge of 244 million dam^3 and annual flow of 7790 m^3/s, which is second only to the Missouri–Mississippi River system. Fifteen percent (100,620 km^2) of the Columbia River Basin (CRB) is located within Canada.

The human population of the Pacific Northwest uses the CR and its tributaries for transportation, irrigation, hydroelectric power, recreation, and as a source of fish for food. Historically, high water quality of the mainstem CR has been attributed to the dilution effect of pristine snowmelt that represents most of the stream flow. However, the CR system supports numerous extractive industries including mining, timber, and commercial fishing. These uses and demands have resulted in listings of impaired waters, fish consumption advisories, and threatened and endangered species. Although federal and state programs have measured contaminants and monitored water quality in the CRB, adverse impacts from environmental contaminants to fish within this system are poorly understood (Schneider, 2002). Mining, agriculture, irrigation, hydroelectric dams, industrial discharges, urban runoff, grazing, and logging have all been associated with degraded water quality in the CRB (Rinella et al., 1993; Joy and Patterson, 1997; Wentz et al., 1998; Williamson et al., 1998; Schneider, 2002). Pesticides are used heavily in agricultural areas including irrigated and dryland farming of the Central Columbia Plateau (Williamson et al., 1998), Willamette Valley (Wentz et al., 1998) and Snake River Basin (Clark and Maret, 1998), where aquatic life may be at risk from pesticides. Industrial discharges from bleached kraft pulp and paper mills, sewage outfalls, and water treatment plants are sources of chlorinated dioxins and furans to the lower CR and Willamette River (Curtis et al., 1993). Mining activities in the upper CRB have contaminated portions of the Clark Fork River in Montana (MT) and the Coeur d'Alene River in Idaho (ID) and have resulted in metal bioaccumulation to hazardous levels in fish and birds (Henny et al., 1994, 2000; Farag et al., 1995, 1998). The U.S. Department of Energy Hanford Site on the CR downstream from Richland, Washington (WA) produced radioactive materials until the early 1990s; high concentrations of PCBs and other organic chemicals have been measured for several decades in fish collected downstream from the facility (USEPA, 2002a). Hydroelectric dams on the CR and Snake River have been associated with declines in commercial stocks of salmon and trout and are also PCB sources.

Native American populations depend heavily on CRB fish as a food source and may be exposed to high concentrations of contaminants from the fish they consume (USEPA, 1992). Risks were primarily attributed to PCBs, *p,p′*-DDE, chlorinated dioxins and furans, arsenic (As), and mercury (Hg), and the USEPA recommended that regulatory agencies should continue to monitor these contaminants (USEPA, 2002a).

We sampled the CR and several of its largest tributaries during fall 1997 and early spring 1998. Our primary objective was to document and assess spatial and temporal trends in the concentrations of environmental contaminants and their effects in CRB fish. Secondary objectives were to compare results from the CRB to other US river systems and to further define benchmarks for the quantification of long-term trends and interpretation of biomarker results. These latter objectives were achieved by building on the results of similar investigations conducted in the Rio Grande Basin (RGB) in 1997 (Schmitt et al., 2005) and the Mississippi River Basin (MRB) in 1995 (Schmitt,

2002a). In this paper, we summarize the most pertinent findings of the CRB study, which are reported in greater detail by Hinck et al. (2004a). Data from this and related Large River Monitoring Network (LRMN) investigations are available at <www.cerc.usgs.gov/data/best/search/index.htm>.

2. Materials and methods

A suite of chemical and biological methods was used to characterize the exposure of fish to contaminants and the effects of exposure. The methods included exposure indicators [concentrations of organochlorine and elemental contaminants, 2,3,7,8-tetrachloro dibenzo-*p*-dioxin equivalents (TCDD-EQ), and hepatic ethoxyresorufin *O*-deethylase (EROD) activity], fish health indicators [ponderal and somatic indices, external lesions, health assessment index (HAI), and general histopathology], and reproductive health indicators [gonadosomatic index (GSI), gonadal histopathology, and plasma vitellogenin (vtg) concentrations]. Detailed descriptions of all procedures and quality assurance (QA) results are presented elsewhere (Schmitt and Dethloff, 2000; Schmitt, 2002b; Hinck et al., 2004a; Schmitt et al., 2005).

2.1. Sampling and field procedures

Fish were collected at 16 sites (Fig. 1; Table 1). Eight were located on the CR in WA and Oregon (OR), two were on the Willamette River in western OR, three were on the Snake River in ID and WA, and one site each were on the Yakima River in WA, Salmon River in ID, and Flathead River in MT. Ten sites (Stations 41, 42, 43, 44, 45, 46, 96, 97, 98, and 117) were National Contaminant Biomonitoring Program (NCBP) stations where contaminants in fish were monitored from the late 1960s to the mid-1980s (Schmitt et al., 1999). These sites were selected to ensure spatial and temporal continuity with historical data and to facilitate trend analysis. Stations 501, 502, 503, 504, and 505 were National Stream Quality Accounting Network (NASQAN) sites (Hooper et al., 2001). One site (Station 506) was not associated with previous monitoring studies. Most fish were collected between early September and November 1997, but Stations 501 and 506 were not sampled until April 1998 (Table 1).

Ten males and 10 females of each target species, largemouth bass (*Micropterus salmoides*) and common carp (*Cyprinus carpio*, henceforth carp), were collected at each site. Alternative preferred species were

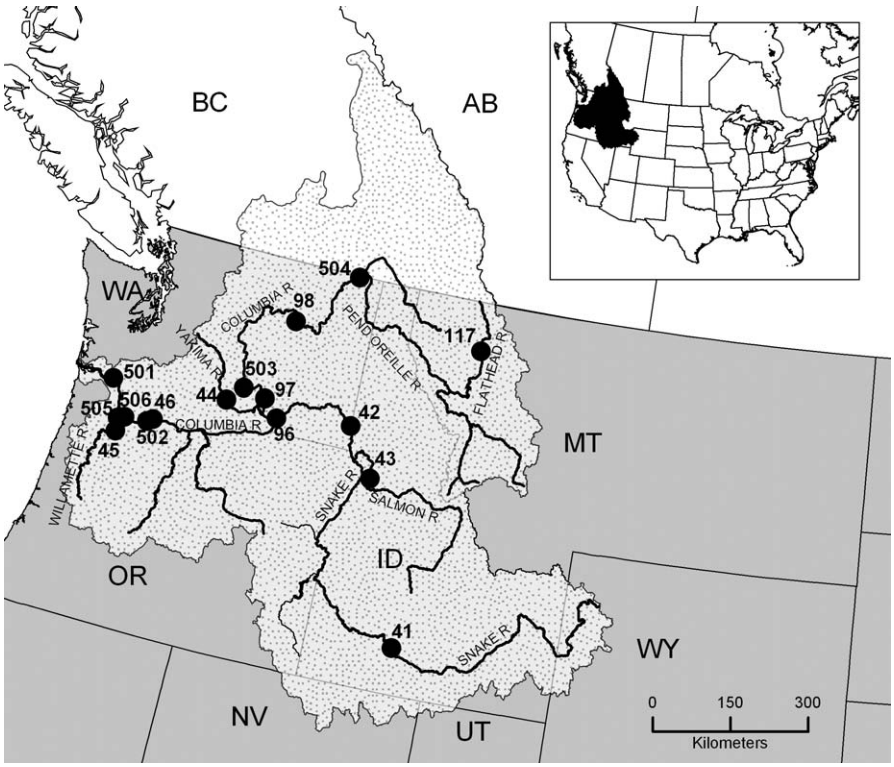

Fig. 1. Map of the Columbia River Basin illustrating waterways, state and international boundaries, and locations sampled.

Table 1
Location, program of origin, and collection dates of sampling stations in the Columbia River Basin (ordered upstream to downstream)

Sub-basin and river	Station number	Location	Collection date(s)	Latitude, longitude
Upper CRB				
Flathead	117 [a]	Creston, MT	10/31/97–11/1/97	48°09′01.09″N, 114°11′29.71″W
Columbia	504 [b]	Northport, WA	11/3/97–11/4/97	48°58′21.70″N, 117°38′48.92″W
Columbia	98 [a]	Grand Coulee, WA	11/6/97–11/7/97	47°57′44.85″N, 118°58′53.84″W
Snake River Basin				
Snake	41 [a]	Hagerman, ID	10/2/97	42°47′36.21″N, 114°56′18.10″W
Salmon	43 [a]	Riggins, ID	9/30/97	45°35′43.42″N, 116°16′55.00″W
Snake	42 [a]	Lewiston, ID	9/24/97–9/25/97	46°24′54.28″N, 117°02′03.49″W
Snake	96 [a]	Ice Harbor Dam, WA	10/8/97–10/9/97	46°41′51.68″N, 118°53′07.88″W
Middle CRB				
Columbia	503 [b]	Vernita Bridge, WA	10/13/97–10/14/97	46°37′28.40″N, 119°51′31.45″W
Columbia	97 [a]	Pasco, WA	10/10/97–10/11/97	46°31′49.22″N, 119°16′42.07″W
Yakima	44 [a]	Granger, WA	10/15/97–10/16/97	46°20′49.31″N, 120°12′27.03″W
Lower CRB				
Columbia	46 [a]	Cascade Locks, OR	11/18/97–11/20/97	45°41′23.11″N, 121°51′00.41″W
Columbia	502 [b]	Warrendale, OR	11/25/97–11/26/97	45°38′00.82″N, 121°58′42.57″W
Columbia	506	Vancouver, WA	4/2/98	45°35′44.21″N, 122°32′13.61″W
Willamette	45 [a]	Oregon City, OR	11/21/97–11/24/97	45°19′03.47″N, 122°39′57.50″W
Willamette	505 [b]	Portland, OR	11/14/97–11/17/97	45°33′04.51″N, 122°41′43.74″W
Columbia	501 [b]	Beaver Army Terminal, OR	3/31/98–4/1/98	46°10′57.86″N, 123°04′13.87″W

[a] National Contaminant Biomonitoring Program (NCBP) site (Schmitt et al., 1999).
[b] National Stream Quality Accounting Network (NASQAN) site (Hooper et al., 2001).

largescale sucker (*Catostomus macrocheilus*) and other black basses (*Micropterus* spp.). More than two species were collected at sites with incomplete quotas for the target or preferred alternate taxa (Hinck et al., 2004a). Adult fish of a size representative of those believed to be present at the sites based on extant information were sought, and extremely large or small fish were avoided.

Fish were collected by boat electrofishing and held in aerated live wells until processed. All collection, handling, and euthanasia procedures followed animal care and use guidelines (American Fisheries Society et al., 2004). A blood sample was obtained from the posterior caudal artery and vein and was chilled on [wet] ice. The fish was then weighed, measured, and killed with a blow to the head. Observations of external features were recorded, and tissue anomalies were dissected and preserved in 10% neutral buffered formalin (NBF) for histopathological analysis. The liver (in species other than carp and sucker), spleen, and gonads were removed and weighed. The liver, gall bladder, posterior and anterior kidneys, gonads, mesenteric fat, and spleen were examined. Pieces of liver were collected and immediately flash-frozen in a dry ice-ethanol slush for EROD analysis. Samples of gonad, kidney, spleen, and additional pieces of liver were collected and preserved for histopathological examination, gender confirmation (gonad), and macrophage aggregate analysis (spleen). Scales were collected for age determination. All remaining tissues (those not frozen or fixed) were wrapped in aluminum foil and frozen for analysis of organochlorine chemical and elemental contaminants and TCDD-EQ. Contact instruments and work surfaces were cleaned between fish to prevent cross-contamination. After blood samples were centrifuged, the plasma was aspirated and frozen in dry ice for vtg analysis. Cryogenically frozen liver and plasma samples were shipped to the laboratory in nitrogen dry shippers and stored at −80 °C. Frozen fish carcasses were shipped in dry ice to the analytical laboratory and stored at −20 °C.

2.2. Laboratory analyses

Composite fish samples were prepared for organochlorine chemical residues, elemental contaminants, and TCDD-EQ analyses by band-sawing and grinding with a commercial meat grinder. The composite sample was then sub-sampled and re-frozen (−20 °C). One subsample (100 g) was freeze-dried, and a portion was acid-digested and analyzed for 19 elemental contaminants by atomic absorption spectroscopy and inductively coupled plasma emission spectroscopy. Moisture content was

determined as weight loss during lyophilization. QA measures for elemental analyses included the analysis of reagent blanks, duplicate samples, certified reference materials, and fortified samples. All elemental concentrations were converted from dry-weight (dw) to wet-weight (ww) concentrations for statistical analysis and reporting using the moisture content of each sample. Limits-of-detection (LODs) were determined individually for each analyte in each sample. The nominal LODs were 0.21 μg/g for As, 0.04 μg/g for cadmium (Cd), 0.05 μg/g for total Hg, 0.09 μg/g for Pb, and 0.22 μg/g for Ni and Se. A second sub-sample (10 g) was solvent-extracted and analyzed gravimetrically for lipid content and by gas chromatography with electron capture detection for 21 organochlorine pesticide residues and total polychlorinated biphenyls (PCBs). QA measures for the organochlorine analyses included the analysis of duplicate and fortified samples and the confirmation of residue identities in selected samples by gas chromatography-mass spectrometry. Recovery efficiency ranged from 88.6% for α-BHC to 99.2% for mirex, but was 90–95% for most analytes. Nominal LODs were 0.01 μg/g for individual compounds and 0.03 μg/g for toxaphene and total PCBs. Residue concentrations were not adjusted for recovery efficiency. A third sub-sample (10 g) was solvent extracted and subjected to reactive cleanup for use in the H4IIE bioassay as previously described (Tillitt et al., 1991; Tysklind et al., 1994). Concentrations of TCDD-EQ (pg/g ww) were determined by slope ratio assay (Finney, 1980) as described by Ankley et al. (1991). QA measures included the analysis of duplicate samples and reference materials. Limits-of-quantification (LOQs; all 1 pg/g) and LODs (0–1 pg/g) were computed separately for each set of samples.

Hepatic EROD activity was determined on microsomal fractions as previously described (Whyte et al., 2000; Hinck et al., 2004a), and protein content was determined using the fluorescamine protein assay (Lorenzen and Kennedy, 1993). EROD activity was reported as the mean of triplicate determinations. LODs were 0–0.15 pmol/min/mg, and LOQs were 0–0.35 pmol/min/mg. Additional QA measures included the analysis of reference materials and duplicate samples.

Preserved tissues were prepared for histopathological analysis as described by Blazer et al. (2002). Paraffin-embedded tissue sections (6 μm) mounted on glass slides were stained with hematoxylin and eosin (H&E) for microscopic examination. Macrophage aggregates (MA) and MA pigments in spleen sections were stained using Perl's method (Luna, 1992). All MA measurements were made with a computer-based image analysis system, and included the number of aggregates in 2 mm^2 of tissue (MA-#) and the mean size (area) of aggregates within those 2 mm^2 (MA-A). The percentage of tissue occupied by aggregates (MA-%) was computed from these measurements. Transverse ovary sections were assigned to developmental stages 0–5 based on the predominant size and appearance of oocytes (Treasurer and Holliday, 1981; Nagahama, 1983; Rodriguez et al., 1995; McDonald et al., 2000; Blazer, 2002); one hundred oocytes in each sample were counted to quantify atresia. Transverse testes sections were similarly classified into developmental stages 0–4 (Nagahama, 1983; Blazer, 2002). Gonadal tissue was also examined microscopically for abnormalities such as intersex, parasites, and neoplasia. Male fish were identified as intersex when individual or small foci of undeveloped oocytes were observed within testicular tissue (i.e., when an ovotestis condition was detected).

Concentrations of vtg in smallmouth (*Micropterus dolomieu*) and largemouth bass (henceforth bass), carp, and largescale sucker were determined by enzyme-linked immunosorbent assay (ELISA; Denslow et al., 1999). The LOD was 0.002 mg/mL for bass, 0.005 mg/mL for carp, and 0.0005 mg/mL for largescale sucker. All assays were performed in triplicate and reported as the mean of triplicate measurements. The coefficient of variation was <10% for all samples analyzed. Inter-assay variability was <10% as determined by routinely analyzing controls on several plates.

2.3. Data set composition and statistical analyses

A total of 560 fish representing eight species and 16 stations were collected and examined. Bass (n=134), carp (n=157), and largescale sucker (n=159) together represented 80% of the total. Bass were collected from 12 stations, largescale sucker from ten, carp from nine, and northern pikeminnow (*Ptychocheilus oregonensis*) from seven. The other three species [longnose sucker (*Catostomus catostomus*), walleye (*Stizostedion vitreum*), and rainbow trout (*Oncorhynchus mykiss*)] were collected from only one or two stations. Composite samples (n=64) were analyzed for organochlorine chemical residues, elemental contaminants, and TCDD-EQ. Of these, 17 samples from nine stations were carp, 17 samples from nine stations were largescale sucker, and 21 samples from 11 stations were bass. The remaining nine samples comprised northern pikeminnow (n=6; five stations), rainbow trout (n=2; one station), and walleye (n=1; one station).

The occurrence of gross external pathological disorders was determined during field processing. To

maintain consistency with previous studies (e.g., Fournie et al., 1996, 2001; Blazer et al., 2002; Schmitt et al., 2005), only the following observations were included: grossly visible disorders of the eye, opercles, body surface, fins, and skeleton. A necropsy-based HAI score was also calculated for each fish by assigning numerical values to gross lesions (Adams et al., 1993; Blazer et al., 2002), then summing the values for all organs observed. An HAI score was computed for a fish only if observations were present for all components.

Body and organ weights were used to compute condition factor (CF) and organosomatic indices (Blazer et al., 2002) according to the following formulae: CF = body weight/length^3; hepatosomatic index (HSI) = liver weight/(total body weight − gonad weight) × 100; splenosomatic index (SSI) = spleen weight/(total body weight − gonad weight) × 100; gonadosomatic index (GSI) = gonad weight/total body weight × 100. The weight of the gonads was subtracted from the body weight to minimize the effect of the reproductive cycle on these indices (Schmitt and Dethloff, 2000).

Bass were grouped at the genus level for statistical analysis. Most biomarker results were analyzed using analysis-of-variance (ANOVA) and analysis of covariance (ANCOVA) to test for differences among sites and to examine effects due to age, gender, and gonadal stage. Least-square means, which are adjusted for all effects in the model, were tested. Transformations were applied to approximate the normality and homogeneity-of-variance required for the application of these parametric statistical methods. Concentrations of contaminants (including TCDD-EQ), EROD activities, and vtg concentrations were $\log_{10}$-transformed; HAI scores were rank-transformed. External lesion frequencies were not analyzed statistically but were accounted for in the HAI scores. All computations and statistical analyses were performed with Version 8 of the Statistical Analysis System (SAS Institute, 1999).

Fish for which only regenerated scales were collected (25 carp, one largemouth bass, seven largescale sucker, nine northern pikeminnow) were excluded from all analyses that included age as a factor. Fish for which the gender could not be verified histologically (including four of the targeted species) were also excluded from analyses that included gender as a factor. Biomarker data for bass, carp, and largescale sucker were summarized and are presented in more detail than other species.

All results for analytes in composite samples were converted to, reported as, and analyzed statistically as ww concentrations. A value of one-half the LOD was substituted for censored values in all statistical analyses and graphs. Concentrations of many contaminants were <LOD, which limited the extent and rigor of statistical analyses that could be performed. Geographic differences in concentrations of As, Cd, Hg, Se, Zn, *p,p′*-DDE, and PCBs were examined statistically using ANOVA, as were temporal differences by combining results of our study with 1969–1986 NCBP data from Stations 41, 42, 43, 44, 45, 46, 96, 97, 98, and 117 (Schmitt et al., 1999). Analytical methods from the NCBP were comparable to those in the present study, although quantification of PCBs differed slightly and LODs were generally lower for contaminants in 1997 (Schmitt et al., 1999). Temporal and spatial differences in $\log_{10}$-transformed length-adjusted (HgL) and weight-adjusted (HgW) Hg concentrations were computed as described by Brumbaugh et al. (2001), and temporal and spatial differences were also tested by ANOVA because concentrations of total Hg in predatory fish increase with size, age, or both (Wiener et al., 2002). A nominal α-level of 0.05 was used in all statistical tests unless otherwise indicated. Details of the statistical procedures are given in Hinck et al. (2004a).

3. Results

3.1. Lipid and moisture content (data not shown)

Lipid content differed among sites and species, but was typically 2–6% lipid. Carp samples generally contained 3–8% lipid except those from Stations 42, 45, and 96, which contained 10.8–15.2%. Samples contained 64–78% water; largescale sucker generally had the highest moisture content (>73%).

3.2. Exposure indicators

3.2.1. Elemental contaminants

Concentrations of As were >LOD (0.21–0.31 μg/g) in 16 samples (25%) from nine stations (Fig. 2). The greatest concentrations (0.30–0.56 μg/g) were in bass from Stations 96 and 502, carp from Stations 44, 96, 97, and 502, and largescale sucker from Stations 43, 501, and 504 (Fig. 2). Differences among stations were statistically significant in northern pikeminnow but not in bass, carp, or largescale sucker (Table 2). Concentrations of As in 1997 differed significantly from historical concentrations in carp from Stations 44, 45, 96, and 97, largescale sucker from Stations 45, 46, and 98, and bass from Stations 43 and 44 (Table 3). Concentrations were stable or increasing at many sites.

Selenium was detected in 55 of 64 samples (86%); concentrations ranged from 0.19 to 1.10 μg/g (Fig. 2).

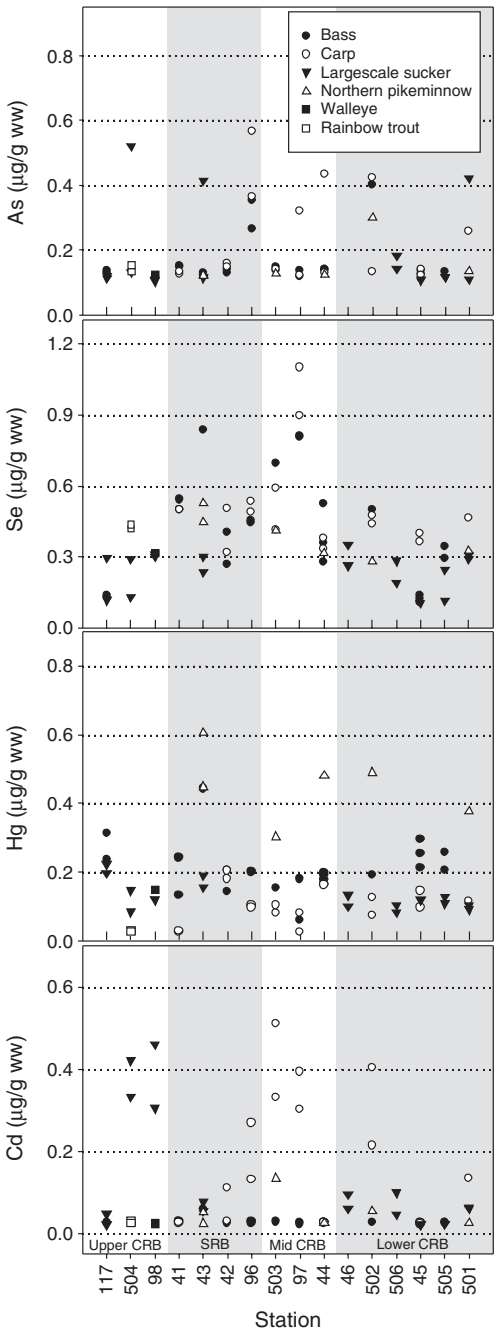

Fig. 2. Concentrations of arsenic (As), selenium (Se), total mercury (Hg), and cadmium (Cd) in composite samples of whole fish, by sub-basin, station, and taxon. (Note: censored values are plotted as 50% of LOD.)

The greatest concentrations (≥0.6 μg/g) were in carp from Station 97 and bass from Stations 43, 97, and 503. Concentrations in largescale sucker were all <0.4 μg/g. Among-station differences were significant in carp and bass but not in largescale sucker or northern pikeminnow (Table 2). Concentrations of Se in 1997 differed significantly from historical concentrations in carp from Stations 42 and 97, largescale sucker from Stations 45, 46, and 98, bass from Station 44, and northern pikeminnow from Station 43, but increasing or decreasing temporal trends were not evident (Table 3).

Concentrations of total Hg were >LOD (0.05 μg/g) in 59 of 64 samples (92%). The greatest concentrations (>0.40 μg/g) were in northern pikeminnow from Stations 43, 44, and 502 and bass from Station 43 (Fig. 2). Concentrations were generally greater in piscivores than in benthivores. Differences in total Hg, HgL, and HgW among stations were significant in bass and carp (Table 2). Temporal differences were significant in some taxa, and these differences were consistent for total Hg, HgL, and HgW (Table 3). Overall, Hg concentrations in carp from Stations 42, 96, and 97, largescale sucker from Stations 43, 45, 46, 98, and 117, and northern pikeminnow from Station 43 differed significantly among years, but marked temporal trends were not evident (Table 3).

Lead concentrations were >LOD (0.09–0.14 μg/g) in 15 of 64 samples (23%) from eight stations. The maximum concentrations were in female largescale sucker from Stations 504 (9.3 μg/g) and 98 (1.6 μg/g) and in male largescale sucker from Station 504 (4.1 μg/g; data not shown). All other concentrations were <0.68 μg/g; all concentrations in bass, northern pikeminnow, and walleye samples were <LOD. Lead was not analyzed statistically because of the many censored values.

Concentrations of Cd were >LOD (0.043–0.063 μg/g) in 27 samples (42%) from 12 stations and were greater in carp and largescale sucker than in other taxa. Eleven samples had concentrations >0.2 μg/g; the maximum concentration (0.51 μg/g) was in female carp from Station 503 (Fig. 2). Among-station differences were significant in carp and largescale sucker but not in bass or northern pikeminnow (Table 2). Concentrations differed significantly from historical concentrations in carp from Stations 42, 44, 96, and 97, largescale sucker from Stations 43, 46, 98, and 117, and bass and northern pikeminnow from Station 43. Increasing or decreasing temporal trends were not evident (Table 3).

Zinc (Zn) was detected in all samples and was generally greater in carp than in other taxa (Table 2). Concentrations ranged from 11.6 to 105.6 μg/g, the latter in female carp from Station 45. Concentrations of Zn differed significantly among stations only in largescale sucker (Table 2). The only significant temporal difference was also in largescale sucker from Station 98 (Table 3).

Table 2
Least-squares geometric mean concentrations [a] (all wet-weight) of arsenic (As), cadmium (Cd), total mercury (Hg), weight-adjusted total Hg (HgW), length-adjusted total Hg (HgL), lead (Pb), selenium (Se), zinc (Zn), *p,p′*-DDE (DDE), and total PCBs (PCB) in composite samples of whole fish

Taxon, station	As μg/g	Cd μg/g	Hg μg/g	HgW μg/g/kg	HgL μg/g/m	Pb μg/g	Se μg/g	Zn μg/g	DDE μg/g	PCB μg/g
Carp										
41 - Hagerman, ID	0.13a	0.03a	0.03a	0.03a	0.07a	0.05a	0.50a	73.4a	0.16a	0.02a
42 - Lewiston, ID	0.15a	0.07ab	0.19c	0.06ab	0.33b	0.06	0.41a	65.3a	0.58bcd	0.15a
96 - Ice Harbor Dam, WA	0.46a	0.20bc	0.10bc	0.03a	0.16a	0.06	0.51ab	65.5a	0.81d	0.24a
503 - Vernita Bridge, WA	0.14a	0.42d	0.09bc	0.04ab	0.16ab	0.27	0.50ab	73.4a	0.83d	0.33a
97 - Pasco, WA	0.22a	0.35d	0.04ab	0.05ab	0.16ab	0.09	1.00c	88.5a	0.27ab	0.04a
44 - Granger, WA	0.28a	0.03a	0.16c	0.10b	0.32ab	0.05	0.36ab	78.4a	0.72cd	0.10a
502 - Warrendale, OR	0.28a	0.31cd	0.10bc	0.04ab	0.18ab	0.05	0.46ab	82.7a	0.31abc	0.14a
45 - Oregon City, OR	0.13a	0.03a	0.12c	0.03a	0.19ab	0.05	0.38b	79.9a	0.10a	0.32a
501 - Beaver Army, OR	0.26a	0.14abc	0.11bc	0.05ab	0.21ab	0.05	0.46ab	82.0a	0.51abcd	0.25a
Largescale sucker										
117 - Creston, MT	0.12a	0.04a	0.21a	0.18a	0.42a	0.05	0.21a	22.8a	0.01a	0.02a
504 - Northport, WA	0.33a	0.38b	0.11a	0.07a	0.21a	6.17	0.21a	48.3b	0.02a	0.17ab
98 - Grand Coulee, WA	0.11a	0.38b	0.12a	0.10a	0.23a	1.04	0.31a	35.4ab	0.01a	0.02a
43 - Riggins, ID	0.27a	0.07a	0.17a	0.19a	0.38a	0.13	0.27a	22.2a	0.20a	0.02a
46 - Ice Harbor Dam, WA	0.12a	0.08a	0.11a	0.09a	0.22a	0.07	0.31a	22.1a	0.31ab	0.11ab
506 - Vancouver, WA	0.16a	0.07a	0.09a	0.11a	0.21a	0.24	0.24a	20.5a	0.14a	0.43b
45 - Oregon City, OR	0.11a	0.02a	0.12a	0.12a	0.27a	0.04	0.11a	18.9a	0.25ab	0.02ab
505 - Portland, OR	0.12a	0.02a	0.12a	0.17a	0.27a	0.10	0.18a	19.0a	0.10a	0.21ab
501 - Beaver Army, OR	0.27a	0.06a	0.10a	0.14a	0.22a	0.04	0.30a	19.3a	0.67b	0.20ab
Bass										
117 - Creston, MT	0.13a	0.03a	0.27bc	0.27bc	0.72bcd	0.05	0.13ab	16.5a	0.01a	0.02a
41 - Hagerman, ID	0.15a	0.03a	0.18abc	0.23abc	0.50abc	0.06	0.52de	17.4a	0.35ab	0.02a
43 - Riggins, ID	0.01a	0.06a	0.44c	1.30e	1.54d	0.05	0.84f	17.3a	0.20ab	0.02ab
42 - Lewiston, ID	0.14a	0.03a	0.17abc	0.75de	0.67bcd	0.05	0.34cd	17.1a	0.11ab	0.35ab
96 - Ice Harbor Dam, WA	0.31a	0.03a	0.14ab	0.18ab	0.39ab	0.06	0.45cd	12.6a	0.23ab	0.03a
503 - Vernita Bridge, WA	0.15a	0.08a	0.15abc	0.09a	0.33ab	0.06	0.70ef	14.4a	0.51b	0.30ab
97 - Pasco, WA	0.13a	0.03a	0.10a	0.16ab	0.31a	0.05	0.81f	16.9a	0.21ab	0.03a
44 - Granger, WA	0.14a	0.03a	0.18abc	0.43cd	0.60abc	0.06	0.42cd	14.9a	0.82c	0.39ab
502 - Warrendale, OR	0.40a	0.03a	0.19abc	0.25abc	0.53abc	0.06	0.50cde	12.4a	0.18ab	0.38ab
45 - Oregon City, OR	0.12a	0.03a	0.25bc	0.46cd	0.77cd	0.05	0.12a	16.0a	0.19ab	0.02a
505 - Portland, OR	0.33a	0.03a	0.23abc	0.50cde	0.75bcd	0.05	0.32bc	14.4a	0.15ab	0.47b
Northern pikeminnow										
43 - Riggins, ID	0.02a	0.03a	0.53a	0.84b	1.35a	0.05	0.49a	20.8a	0.09a	0.05a
503 - Vernita Bridge, WA	0.65b	0.14a	0.30a	0.29a	0.62a	0.05	0.41a	19.6a	0.28ab	1.30c
44 - Granger, WA	0.09a	0.04a	0.48a	0.80ab	1.16a	0.05	0.32a	16.0a	0.80b	0.17ab
502 - Warrendale, OR	0.30ab	0.05a	0.49a	0.42ab	0.95a	0.05	0.28a	17.1a	0.37ab	0.68b
501 - Beaver Army, OR	0.19ab	0.10a	0.38a	0.46ab	0.89a	0.05	0.33a	17.6a	0.22ab	0.38ab
ANOVA										
F	1.55	10.5*	12.72	17.9*	10.9*	ND [b]	13.9*	14.5*	6.09*	3.71*
df	33, 26	33, 26	33, 26	32, 24	32, 24	ND [b]	33, 26	33, 26	33, 26	33, 26

Within each taxon-station group, means followed by the same letter are not significantly different ($P<0.01$, Fisher's protected LSD). Also shown are ANOVA *F*-values, degrees-of-freedom (*df*), and statistical significance (*, $0.01<P\leq0.05$; **, $P\leq0.01$). ND, not measured.

[a] Censored values (i.e., <LOD) were represented by 50% of the LOD in all computations.

[b] Pb not tested statistically due to large numbers of censored (i.e. <LOD) values.

3.2.2. Organochlorine pesticides

Concentrations of *p,p′*-DDT, the active ingredient in commercial DDT, were >LOD (0.01 μg/g) in 13 of 64 samples (data not shown). However, *p,p′*-DDE, the most persistent metabolite of *p,p′*-DDT, was detected in 60 of 64 samples representing all stations except Station 117 (Fig. 3). Concentrations of *p,p′*-DDE were greatest in carp from Station 42 (0.56–0.59 μg/g); carp, bass, and northern pikeminnow from Station 44 (0.50–1.2 μg/g); carp from Station 96 (0.70–0.92 μg/g); carp and

Table 3
Least-squares geometric mean concentrations [a] (all wet-weight) of arsenic (As), cadmium (Cd), total mercury (Hg), length-adjusted total Hg (HgL), weight-adjusted total Hg (HgW), lead (Pb), zinc (Zn), selenium (Se), *p,p′*-DDE (DDE), and total PCBs (PCB) in fish collected from 1969 to 1997 at National Contaminant Biomonitoring Program (NCBP) sites (Schmitt et al., 1999) in the Columbia River Basin (ordered upstream to downstream)

Taxon, station, location	Year	As μg/g	Cd μg/g	Hg μg/g	HgW μg/g/kg	HgL μg/g/m	Se μg/g	Pb μg/g	Zn μg/g	DDE μg/g	PCB μg/g
Carp											
41 - Hagerman, ID	1974	ND	ND	ND	ND	ND	ND	ND	ND	0.31	ND
	1997	0.13	0.03	0.03	0.03	0.07	0.50	0.05	73.3	0.16	0.02
42 - Lewiston, ID	1970	ND	ND	0.25	0.24**	0.64	ND	ND	ND	0.47	ND
	1971	0.07	0.05	0.30	0.30**	0.80*	ND	0.11	ND	0.16*	ND
	1972	0.14	0.88**	0.17	0.12	0.39	0.17**	0.05	ND	1.10	ND
	1973	0.18	0.06	0.06*	0.05	0.14*	0.30	0.05	ND	0.31	ND
	1997	0.15	0.06	0.19	0.06	0.38	0.40	0.06	65.3	0.58	0.15
96 -Ice Harbor Dam, WA	1971	0.11**	0.07*	0.21*	0.29**	0.66**	ND	0.07	ND	0.41	ND
	1997	0.45	0.19	0.10	0.03	0.16	0.51	0.06	65.2	0.80	0.23
97 - Pasco, WA	1970	ND	ND	0.07	0.12**	0.22*	ND	ND	ND	1.12*	ND
	1971	0.04**	0.10**	0.08	0.13**	0.24**	ND	0.37	ND	0.33	ND
	1972	0.12	1.80**	0.12*	0.27**	0.39**	0.40**	0.20	ND	0.67	ND
	1973	0.18	0.03**	0.01**	0.01	0.02**	0.67	0.09	ND	0.26	ND
	1974	ND	ND	ND	ND	ND	ND	ND	ND	1.54**	ND
	1976	0.30	0.05**	0.02*	0.06*	0.07	ND	0.21	ND	0.14	0.15**
	1980	0.07*	0.05**	0.05	0.09**	0.14	0.83	0.29	78.8	0.19	0.22**
	1997	0.20	0.35	0.05	0.03	0.09	0.99	0.09	87.0	0.25	0.03
44 - Granger, WA	1970	ND	ND	0.23	0.48**	0.81*	ND	ND	ND	1.23	ND
	1971	0.04**	0.03	0.17	0.33**	0.57	ND	0.05	ND	1.05	ND
	1972	0.20	0.42**	0.24	0.66**	0.90*	0.40	0.05	ND	2.20	ND
	1973	0.03**	0.03	0.07	0.13	0.21	0.22	0.05	ND	0.52	ND
	1974	ND	ND	ND	ND	ND	ND	ND	ND	2.61*	ND
	1978	0.17	0.05	0.22	0.43**	0.63	0.54	0.19	92.5	1.22	0.52**
	1997	0.24	0.03	0.16	0.10	0.32	0.36	0.05	78.3	0.67	0.08
45 - Oregon City, OR	1970	ND	ND	0.17	0.08	0.35	ND	ND	ND	0.34	ND
	1973	0.03**	0.03	0.15	0.25**	0.41	0.18	0.05	ND	0.35	ND
	1974	ND	ND	ND	ND	ND	ND	ND	ND	0.88**	ND
	1997	0.13	0.03	0.12	0.03	0.19	0.38	0.05	75.6	0.10	0.31
Largescale sucker											
117 - Creston, MT	1980	0.06	0.01*	0.10*	0.17	0.25	0.20	0.10	18.2	0.03**	0.20**
	1984	0.07	0.02	0.17	0.17	0.40	0.18	0.06	20.1	0.01	0.17**
	1986	0.12	0.02	0.18	0.16	0.37	0.17	0.10	19.7	0.01	0.08**
	1997	0.12	0.03	0.21	0.18	0.43	0.19	0.05	22.8	0.01	0.02
98 - Grand Coulee, WA	1971	0.16	0.11**	0.05*	0.10	0.15	ND	0.72	ND	0.05*	ND
	1976	0.30	0.33	0.02**	0.03**	0.05**	ND	2.57	ND	0.02	0.50**
	1978	0.20	0.36	0.07	0.08	0.15	0.22	1.12	55.4**	0.04*	0.47**
	1980	0.28*	0.22	0.13	0.15	0.29	0.20*	0.65	28.1	0.06**	0.27**
	1984	0.14	0.09**	0.09	0.21	0.27	0.22	0.25	21.1**	0.02	0.25**
	1997	0.11	0.38	0.12	0.10	0.23	0.31	1.04	35.4	0.01	0.02
43 - Riggins, ID	1969	ND	ND	0.23	0.32	0.60	ND	ND	ND	0.14	ND
	1970	ND	ND	0.41*	0.39	0.91*	ND	ND	ND	0.11	ND
	1971	0.13	0.07	0.26	0.33	0.66	ND	0.08	ND	0.23	ND
	1972	0.13	0.18*	0.11	0.20	0.32	0.22	0.14	ND	0.10	ND
	1973	0.20	0.03	0.15	0.18	0.34	0.32	0.05	ND	0.12	ND
	1974	ND	ND	ND	ND	ND	ND	ND	ND	0.18	ND
	1976	0.50	0.12	0.19	0.25	0.41	ND	0.23	ND	0.13	0.19**
	1984	0.11	0.02**	0.10	0.21	0.27	0.30	0.15	21.0	0.01**	0.20**
	1986	0.11	0.05	0.13	0.17	0.29	0.26	0.18	19.1	0.01**	0.08**
	1997	0.22	0.07	0.17	0.19	0.37	0.27	0.13	22.1	0.19	0.02
46 - Cascade Locks, OR	1969	ND	ND	0.27	0.30*	0.64*	ND	ND	ND	0.36	ND
	1970	ND	ND	0.21	0.23	0.49	ND	ND	ND	0.22	ND
	1972	0.03**	0.16	0.23	0.30*	0.57*	0.14**	0.10	ND	0.47	ND
	1973	0.05*	0.08	0.32**	0.38**	0.71**	0.09**	0.35	ND	0.25	ND

(continued on next page)

Table 3 (*continued*)

Taxon, station, location	Year	As μg/g	Cd μg/g	Hg μg/g	HgW μg/g/kg	HgL μg/g/m	Se μg/g	Pb μg/g	Zn μg/g	DDE μg/g	PCB μg/g
	1974	ND	ND	ND	ND	ND	ND	ND	ND	0.20	ND
	1976	0.87**	0.15	0.05	0.14	0.16	ND	0.10	ND	0.13	1.31**
	1978	0.32*	0.06	0.07	0.09	0.17	0.42	0.25	22.5	0.28	0.37*
	1980	0.43**	0.03*	0.07	0.08	0.16	0.26	0.10	18.3	0.54	0.34*
	1984	0.22	0.04	0.10	0.11	0.23	0.24	0.09	20.8	0.73	0.55**
	1997	0.12	0.08	0.12	0.10	0.23	0.30	0.07	21.9	0.30	0.10
45 - Oregon City, OR	1969	ND	ND	0.18	0.33	0.51	ND	ND	ND	0.15	ND
	1970	ND	ND	0.35*	0.44**	0.88**	ND	ND	ND	0.60	ND
	1971	0.05	0.03	0.30*	0.37*	0.74*	ND	0.05	ND	0.25	ND
	1972	0.06	0.02	0.10	0.18	0.30	0.10	0.10	ND	0.45	ND
	1973	0.03**	0.03	0.13	0.25	0.45	0.07	0.05	ND	0.26	ND
	1974	ND	ND	ND	ND	ND	ND	ND	ND	0.27	ND
	1980	0.07	0.01	0.19	0.16	0.26	0.21*	0.14	22.5	0.18	0.92**
	1997	0.11	0.02	0.12	0.12	0.27	0.11	0.04	18.9	0.25	0.02
Bass											
43 - Riggins, ID	1971	0.03**	0.03	0.36	0.79**	1.25	ND	0.05	ND	0.14	ND
	1972	0.03**	0.03	0.21	0.77**	0.86	0.68	0.05	ND	0.36	ND
	1973	0.05	0.03	0.22	0.35**	0.66	0.83	0.05	ND	0.14	ND
	1974	ND	ND	ND	ND	ND	ND	ND	ND	0.05	ND
	1986	0.02**	0.01**	0.25	0.39	0.73	0.61	0.17	13.0	0.06	0.08*
	1997	0.13	0.06	0.44	1.30	1.54	0.84	0.05	17.3	0.20	0.02
42 - Lewiston, ID	1969	ND	ND	0.15	1.10	0.84	ND	ND	ND	0.30	ND
	1970	ND	ND	0.21	0.58	0.75	ND	ND	ND	0.33	ND
	1971	0.06	0.03	0.28	0.70	1.02	ND	0.05	ND	0.61**	ND
	1978	0.05	0.01	0.19	0.32	0.56	0.44	0.10	15.4	0.28	0.35
	1997	0.14	0.03	0.17	0.76	0.67	0.33	0.05	17.0	0.11	0.18
44 - Granger, WA	1969	ND	ND	0.14	0.34	0.55	ND	ND	ND	0.94	ND
	1970	ND	ND	0.27	0.43	0.86	ND	ND	ND	1.66	ND
	1973	0.03**	0.03	0.18	0.23	0.51	0.10**	0.05	ND	0.77	ND
	1974	ND	ND	ND	ND	ND	ND	ND	ND	1.40	ND
	1997	0.14	0.03	0.19	0.34	0.58	0.38	0.06	15.3	0.82	0.22
45 - Oregon City, OR	1976	0.25	0.05	0.13	0.57	0.56	ND	0.12	ND	0.06	0.65**
	1997	0.12	0.03	0.25	0.46	0.77	0.12	0.05	16.0	0.19	0.02
Northern pikeminnow											
43 - Riggins, ID	1970	ND	ND	1.70**	3.75**	5.11**	ND	ND	ND	0.74**	ND
	1971	0.07	0.03	0.71	1.84*	2.24	ND	0.05	ND	0.31*	ND
	1972	0.07	0.03	0.18*	1.32	0.76	0.27*	0.05	ND	0.22	ND
	1978	0.16	0.01*	0.22	0.35	0.57	0.34	0.10	20.5	0.52*	0.90**
	1980	0.05	0.02	0.26	5.73**	1.55	0.58	0.10	22.1	0.02*	0.15
	1997	0.12	0.04	0.52	0.83	1.35	0.48	0.05	20.8	0.09	0.05
44 - Granger, WA	1972	0.10	0.05	0.64	2.82*	2.33	0.40	0.050	ND	2.70	ND
	1997	0.13	0.03	0.48	0.80	1.16	0.32	0.050	16.1	0.80	0.17
ANOVA-*F*	–	5.45	12.08	10.61	11.45	10.90	10.11	ND [b]	31.49	7.19	9.05
df	–	156, 93	156, 93	186, 100	186, 100	186, 100	123, 70	ND [b]	80, 56	209, 112	93, 64

Within each taxon-station group, values followed by asterisks (*) differ significantly (*, $0.01 < P \leq 0.05$; **, $P \leq 0.01$, Fisher's protected LSD) from 1997 means. Also shown are ANOVA *F*-values, degrees-of-freedom (*df*), and statistical significance (**, $P \leq 0.01$). ND, no data/not measured. –, not applicable.

[a] Censored values (i.e., <LOD) were represented by 50% of the LOD in all computations.

[b] Pb not tested statistically due to large numbers of censored (i.e. <LOD) values.

largescale sucker from Station 501 (0.51–0.68 μg/g); and carp and bass from Station 503 (0.51–1.1 μg/g). Concentrations of *p,p′*-DDD (TDE) were detected in 43 of 64 samples, and relatively high concentrations (0.1–0.2 μg/g) were measured in samples from Stations 96 and 503 (data not shown). Trace concentrations of *o,p′*-DDT (0.01–0.043 μg/g) were detected in samples from Stations 503, 505, and 506; however, only one sample (largescale sucker from Station 506) contained trace concentrations of *o,p′*-DDE and *o,p′*-DDD (0.01 μg/g

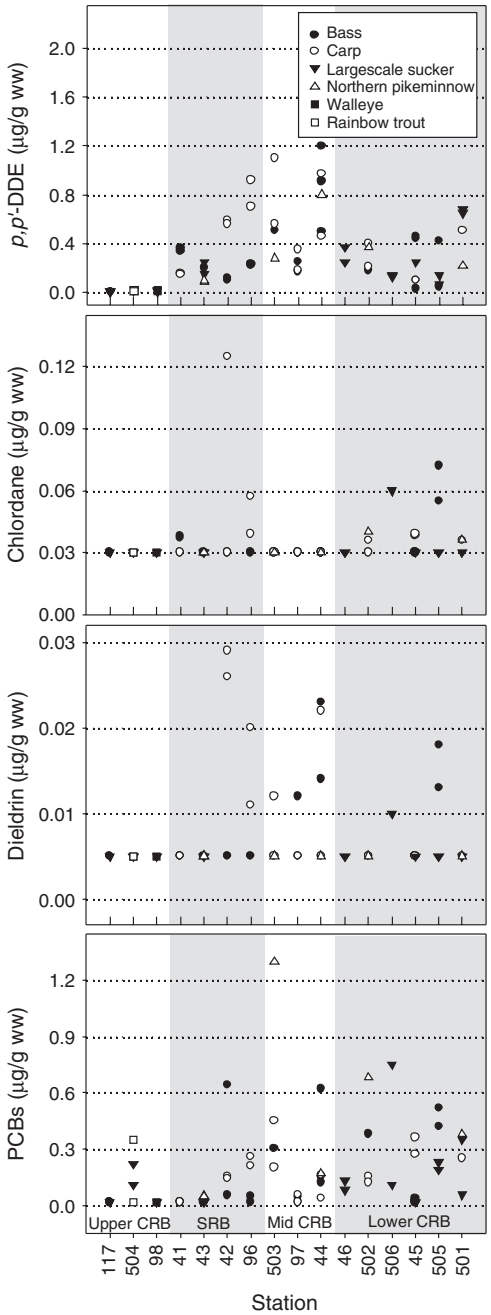

Fig. 3. Concentrations of *p,p′*-DDE, total chlordanes (sum of *cis*- and *trans*-chlordanes and nonachlors, oxychlordane, and heptachlor epoxide), dieldrin, and total PCBS in composite samples of whole fish, by sub-basin, station, and taxon. (Note: censored values are plotted as 50% of LOD.)

for each; data not shown). Among-station differences in *p,p′*-DDE concentrations were significant in all taxa (Table 2). Concentrations differed significantly from historical NCBP concentrations in carp from Stations 42, 44, 45, and 97, largescale sucker from Stations 43, 98, and 117, bass from Station 42, and northern pikeminnow from Station 43; concentrations decreased at most stations (Table 3).

Six chlordane-related compounds (*cis*-chlordane, *trans*-chlordane, *cis*-nonachlor, *trans*-nonachlor, oxychlordane, and heptachlor epoxide) were measured. Of these, *trans*-nonachlor was the most frequently detected constituent and was >LOD (0.01 µg/g) in 15 of 64 samples from eight stations. Concentrations ranged from 0.01 to 0.10 µg/g with the maximum measured in male carp from Station 42 (data not shown). *Cis*-chlordane was >LOD (0.01 µg/g) in female bass from Station 505 (0.02 µg/g), female carp from Station 96 (0.02 µg/g), and male and female largescale sucker from Station 506 (0.01 µg/g; data not shown). Concentrations of *trans*-chlordane, *cis*-nonachlor, oxychlordane, and heptachlor epoxide were <LOD (0.01 µg/g) in all samples except largescale sucker from Station 506, which contained trace concentrations of all four compounds (data not shown). Total chlordane concentrations (sum of six compounds) ranged from 0.03 to 0.13 µg/g and were >0.05 µg/g in carp from Stations 42 and 96, largescale sucker from Station 506, and bass from Station 505 (Fig. 3).

Concentrations of dieldrin were ≥LOD (0.01 µg/g) in 13 of 64 samples from seven stations (Fig. 3). The maximum concentrations were measured in male and female carp (0.029 and 0.026 µg/g, respectively) from Station 42 (Fig. 3). Trace concentrations (0.01–0.023 µg/g) were detected in samples from Stations 44, 96, 97, 503, and 505.

Toxaphene was >LOD (0.03 µg/g) in only two largescale sucker samples, both from Station 506 near Vancouver, WA (data not shown).

3.2.3. Total PCBs and TCDD-EQ

Concentrations of total PCBs were >LOD (0.03 µg/g) in 43 of 64 samples (67%) from 13 stations (Fig. 3). Concentrations ranged from 0.03 to 1.3 µg/g, with the maximum measured in female northern pikeminnow from Station 503. Other samples with concentrations ≥0.5 µg/g included male bass from Station 42 (0.64 µg/g), female bass from Station 44 (0.62 µg/g), female northern pikeminnow from Station 502 (0.68 µg/g), male bass from Station 505 (0.52 µg/g), and female largescale sucker from Station 506 (0.75 µg/g). Among-station differences were significant in largescale sucker, bass, and northern pikeminnow but not in carp (Table 2). The greatest PCB concentrations were generally in fish from the lower CRB (Fig. 3; Table 2). Concentrations were significantly less than historical NCBP concentrations in

carp from Stations 44 and 97, largescale sucker from Stations 43, 45, 46, 98, and 117, bass from Stations 43 and 45, and northern pikeminnow from Station 43 (Table 3). Total PCB concentrations declined in fish from all of these historical sampling sites.

TCDD-EQ concentrations were relatively low in most samples and ranged from <0.68 to 7.0 pg/g in bass, <1.33 to 43 pg/g in carp, and <0.35 to 10 pg/g in largescale sucker (data not shown). TCDD-EQs were >5 pg/g in samples from all sites except Stations 41, 42, 43, 44, 98, and 117. The maximum TCDD-EQs were measured in female carp (43 pg/g) from Station 96 and female largescale sucker (10 pg/g) from Station 46. TCDD-EQs were ≤7 pg/g in other samples from Station 96. TCDD-EQs were generally low (<8 pg/g) in rainbow trout, walleye, and northern pikeminnow.

3.2.4. Hepatic ethoxyresorufin O-deethylase (EROD) activity

Hepatic EROD activity was analyzed statistically only in bass, carp, and largescale sucker because sample sizes for other species were small. In bass, a significant ANOVA model containing the factors station, gender, gonadal stage, and their interactions explained 49% of the total variation in EROD activity ($F_{37,\ 92}=2.41$, $P<0.01$). Differences between genders were significant ($F_{1,\ 92}=4.51$, $P<0.05$); activity was greater in males than in females. The influence of gonadal stage on EROD activity differed between genders ($F_{1,\ 92}=4.21$, $P<0.05$); most fish with EROD activity >20 pmol/min/mg were identified as stage-1 females and stage-2 or -3 males. Hepatic EROD activity also differed among stations. After accounting for all other factors, activity was greatest in bass from Station 505 and lowest in those from Station 503 (Table 4). Mean EROD activity was >15 pmol/min/mg at Stations 42, 43, 117, and 505 in females and Stations 42, 43, 45, 97, 117, 502, and 505 in males.

Hepatic EROD activity in carp was generally lower than in bass. In carp, a significant ANOVA model explained 59% of the total variation in EROD activity ($F_{25,\ 125}=7.28$, $P<0.01$). Genders were analyzed separately even though the main effect was not significant ($F_{1,\ 125}=0.50$, $P>0.05$). Mean EROD activity was <5 pmol/min/mg at most sites (Table 4). Activity in individual fish was >10 pmol/min/mg in males from Stations 42, 44, 45, 96, 502, and 503 and females from Stations 45 and 502.

Hepatic EROD activity in largescale sucker was greater than in carp but similar to bass. Activity did not differ significantly among stations ($F_{22,\ 97}=1.06$, $P>0.05$). However, mean EROD activity differed significantly in female and male largescale sucker (Table 4). Mean EROD activity ranged from 7.23 to 30.7 pmol/min/mg and was lowest in females from Station 504 and males from Station 117 (Table 4). Mean EROD activity was >20 pmol/min/mg in female bass from Stations 98, 502, and 505 and male bass from Station 43 (Table 4). Similar to bass, mean EROD activity was greatest in female largescale sucker from Station 505. Hepatic EROD activity was also measured in longnose sucker, northern pikeminnow, rainbow trout, and walleye (Table 4). Mean EROD activity was >9 pmol/min/mg in northern pikeminnow from Stations 46 and 503, rainbow trout from Station 504 walleye from Station 98.

3.3. Fish health indicators

3.3.1. External lesions and health assessment index (HAI)

Seventy-four percent of CRB fish examined had some type of external lesion, most of which were identified as eroded, frayed, hemorrhagic, or embolic fins. Lesion frequencies for all species combined ranged from 50% at Station 98 to 100% at Station 44 (data not shown). External lesions were identified on 92 of 134 (69%) bass examined. Lesion occurrence for bass was lowest (<45%) at Stations 42, 43, and 503. Of the 157 carp examined, 83% had external lesions. Station 97 had the lowest percentage of carp with external lesions (50%); occurrence at all other stations was >75%. External lesions were identified on 78% of the 160 largescale sucker examined; all stations had >60% lesion occurrence.

In general, fish with high HAI scores were considered to be in poorer health than those with low HAI scores. Most HAI scores for bass (80%) ranged from 0 to 100. Mean HAI scores were <65 except at Stations 41, 44, and 45, which had means >80 (Fig. 4). Most HAI scores for carp (82%) ranged from 0 to 70, which indicated that most fish had zero to three lesions (Fig. 4). Mean HAI scores ranged from 23 to 64, and individual carp from Stations 44, 96, 502, and 503 had HAI scores >110. Most HAI scores for largescale sucker (95%) were 0–100, and means ranged from 30 to 63 (Fig. 4). All largescale sucker from Stations 43, 117, and 502 had HAI scores <70, but two or more fish had HAI scores ≥100 at the other stations.

Histopathological examinations revealed that most external lesions, as well as most nodules or discolorations of liver, kidney, and spleen tissues, were inflammatory responses associated with parasites. Frayed or hemorrhagic fins and liver abnormalities in

Table 4
Geometric means[a] and ranges of hepatic ethoxyresorufin *O*-deethylase (EROD) activity (pmol/min/mg protein) by taxon, station, and gender

Taxon and station	Female			Male			Juvenile[b]		
	n	Range	Mean	*n*	Range	Mean	*n*	Range	Mean
Bass									
117 - Creston, MT	11	11.5–33.8	18.5c	10	14.2–51.8	27.8c	0	–	–
41 - Hagerman, ID	9	3.89–15.8	8.52ab	7	4.32–15.9	7.75a	0	–	–
43 - Riggins, ID	3	5.68–233	21.8cd	4	4.82–34.0	16.4abc	0	–	–
42 - Lewiston, ID	5	10.0–32.9	16.3bc	7	10.9–31.8	18.1bc	1	–	43.1
96 - Ice Harbor Dam, WA	4	6.87–17.2	10.9abc	3	2.30–18.1	8.2ab	0	–	–
503 - Vernita Bridge, WA	3	3.60–7.61	5.21a	2	4.76–8.27	6.28a	0	–	–
97 - Pasco, WA	9	3.15–23.8	11.3abc	6	10.2–29.7	15.2abc	0	–	–
44 - Granger, WA	9	4.3–33.9	10.9abc	6	0.18–36.7	6.9abc	0	–	–
502 - Warrendale, OR	1	–	11.6abcd	3	11.0–22.1	15.8abc	0	–	–
45 - Oregon City, OR	15	2.65–42.2	12.5bc	5	10.1–82.8	29.4cd	1	–	48.8
505 - Portland, OR	5	30.2–57.4	40.4d	3	47.6–99.4	68.3d	0	–	–
ANOVA		$F_{10, 63}=3.00$**			$F_{10, 44}=4.96$**				
Carp									
41 - Hagerman, ID	10	0.58–8.77	2.01bc	10	1.35–4.66	2.52ab	0	–	–
42 - Lewiston, ID	5	0.18–6.66	2.11c	12	0.18–11.5	3.31cd	0	–	–
96 - Ice Harbor Dam, WA	10	0.88–5.72	2.44bc	11	2.35–24.4	8.76d	0	–	–
503 - Vernita Bridge, WA	10	1.45–8.24	3.01c	11	3.88–212	10.6d	0	–	–
97 - Pasco, WA	11	0.05–1.43	0.27a	9	0.06–4.32	0.89a	0	–	–
44 - Granger, WA	10	0.85–3.02	1.62b	10	0.89–25.1	3.00bc	0	–	–
502 - Warrendale, OR	10	1.64–10.8	3.17c	10	3.60–51.7	6.82d	0	–	–
45 - Oregon City, OR	4	6.54–22.9	10.3d	10	0.98–19.0	7.82d	0	–	–
ANOVA		$F_{7, 58}=13.8$**			$F_{7, 72}=6.85$**				
Largescale sucker									
117 - Creston, MT	4	5.27–151	16.2abc	6	0.60–20.0	9.36a	0	–	–
504 - Northport, WA	10	4.00–12.8	7.23a	10	9.53–42.6	19.5ab	0	–	–
98 - Grand Coulee, WA	10	9.07–67.8	22.0bc	10	6.00–59.8	19.7ab	0	–	–
43 - Riggins, ID	11	8.43–33.5	15.7b	9	5.39–86.5	30.5bc	0	–	–
46 - Cascade Locks, OR	11	9.49–48.7	19.5bc	9	0.99–108	14.1ab	1	–	9.31
502 - Warrendale, OR	3	3.50–60.7	23.0bc	0	–	–	0	–	–
45 - Oregon City, OR	6	8.35–24.8	15.6bc	0	–	–	0	–	–
505 - Portland, OR	11	8.45–107	30.7c	10	0.04–107	13.2c	2	–	28.2
ANOVA		$F_{7, 58}=3.35$**			$F_{5, 46}=3.27$*				
Longnose sucker									
117 - Creston, MT	10	2.36–11.0	5.05	1	–	18.0	2	4.12–6.57	5.20
98 - Grand Coulee, WA	1	–	3.00	1	–	7.34	0	–	–
Northern pikeminnow									
43 - Riggins, ID	9	0.18–4.33	1.31	5	0.18–7.79	2.22	0	–	–
503 - Vernita Bridge, WA	8	2.10–102	10.7	2	2.07–26.1	7.4	0	–	–
44 - Granger, WA	5	0.18–3.37	0.78	1	–	0.18	0	–	–
46 - Cascade Locks, OR	2	3.11–48.1	12.2	0	–	–	0	–	–
502 - Warrendale, OR	11	0.64–5.53	2.14	0	–	–	0	–	–
505 - Portland, OR	2	1.59–2.44	1.97	0	–	–	0	–	–
Rainbow trout									
504 - Northport, WA	10	1.19–25.2	9.18	2	6.85–12.0	9.05	8	4.94–41.5	15.1
Walleye									
98 - Grand Coulee, WA	8	9.45–51.8	24.3	4	16.7–26.3	19.5	4	25.8–30.7	28.3
505 - Portland, OR	1	–	4.83	0	–	–	0	–	–

Stations are order upstream to downstream. Within each taxon-gender group for bass, carp, and largescale sucker, means followed by the same letter are not significantly different ($P>0.05$, Fisher's protected LSD). –, not applicable.

[a] Censored values were represented by 50% of the limit-of-quantification in the computation of geometric means.

[b] May include fish of undetermined gender from which no gonad sample was obtained.

bass from Stations 41, 44, and 45 were the main contributors to high HAI scores; liver, spleen, and kidney abnormalities were associated with helminth and myxosporidian parasites. Four bass from Station 41 had nodules of hyperplastic interrenal tissue within the anterior kidney, which may be indicative of chronic

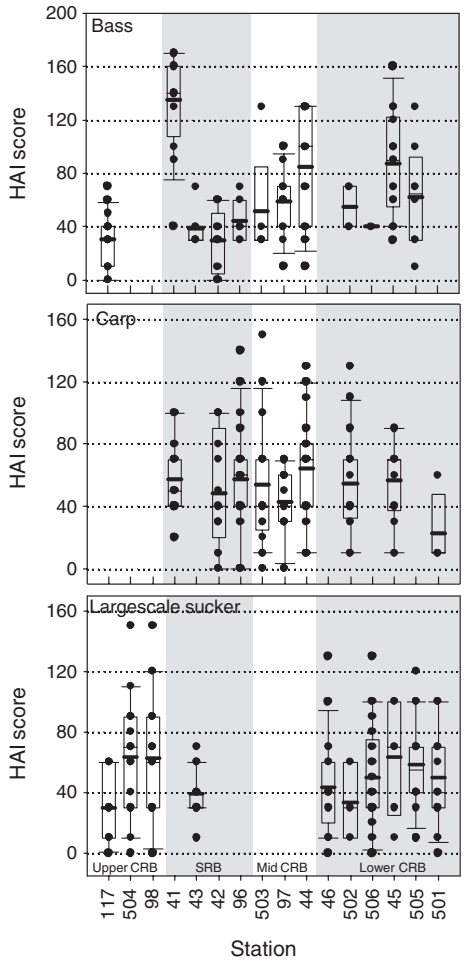

Fig. 4. Health assessment index (HAI) scores of bass, carp, and largescale sucker, by sub-basin and station. Shown for each group are points representing individual fish and the mean (heavy horizontal line), median (light horizontal line), interquartile range (box), and the 10th and 90th percentiles (whiskers). Stations are ordered from upstream to downstream and are grouped by sub-basin.

stress (Fig. 5A). Little or no observable interrenal tissue is typically found in the anterior kidney (Fig. 5B). Testicular tissue in two male carp from Stations 502 and 503 was diagnosed as abnormal. The abnormal testicular tissue at Station 502 was a proliferation of fibrous tissue and inflammation. At Station 503, a tumor of multiple cell origin with components of a leiomyoma (neoplasia of smooth muscle) and a seminoma (neoplasia of germ cell origin) was observed (Fig. 5C and D). Most largescale sucker (85%) from Station 504 had external lesions on the body surface and fins. Five of these lesions were diagnosed as papillomas; two had thickened areas of epithelium with accumulations of pigmented macrophages around vessels in the dermis. Another was an eye lesion composed of chronic granulomatosis inflammation throughout the choroids, iris, and retina. These lesions were similar to those previously described for *Streptococcus* and *Staphylococcus* infections of fish (Shah and Tyagi, 1986; Chang and Plumb, 1996). Thirteen largescale sucker from Station 98 had grossly visible eye abnormalities, with most identified in the field as being opaque. Histologically, the eye lesions were composed of thickened and sometimes inflamed cornea (Fig. 5E) and chronic inflammation of the iris, lens capsule, choroid gland and retina (Fig. 5F).

3.3.2. Condition factor and organosomatic indices (data not shown)

Condition factor, HSI, and SSI were computed from body and organ weights and used as general indicators of overall fish health. Condition factor values of 1.0–2.0 are common in many fish species. A significant ANOVA model containing the factors station, gender, gonadal stage, and their interactions explained 50% of the total variation in condition factor for bass ($F_{38,\ 93}=2.40$, $P<0.01$). Differences among stations were significant ($F_{8,\ 93}=3.95$, $P<0.01$) as were gender differences ($F_{1,\ 93}=4.46$, $P<0.05$). Mean CF values ranged from 1.3 at Station 42 to 2.1 at Station 502 in female bass and 1.3 at Station 42 to 1.8 at Station 96 in male bass; most individual CF values (89%) were between 1.1 and 1.9. Condition factor did not differ significantly in carp ($F_{27,\ 127}=1.37$, $P>0.05$). Mean CF values ranged from 1.2 at Station 44 to 1.9 at Station 42, and all individual values were between 0.9 and 1.9. Similar to bass, the ANOVA model for CF in largescale sucker was statistically significant and explained 48% of the total variation ($F_{28,\ 128}=4.25$, $P<0.01$), and differences among stations were significant ($F_{9,\ 128}=3.44$, $P<0.01$). Mean CF values ranged from 0.86 at Station 98 to 1.2 at Station 45, and greater CF values characterized largescale sucker from Station 45 than those from other stations.

In bass, HSI differences were significant and explained 43% of the total variation ($F_{22,\ 108}=3.66$, $P<0.01$). Differences among stations were also significant ($F_{11,\ 108}=4.17$, $P<0.01$). Station means ranged from 1.0% at Station 502 to 2.3% at Station 503 in female bass and 0.9% at Station 97 to 2.0% at Station 43 in male bass. Most individual values were 0.8–2.4%, but female bass from Stations 43, 45, 97, and 503 and male bass from Stations 43, 44, and 117 had HSI values >2.1%. All female bass from Station 503 had HSI values >2.0%.

Differences in SSI values in bass were not significant ($F_{22,\ 107}=1.45$, $P>0.05$). Mean SSI values ranged from

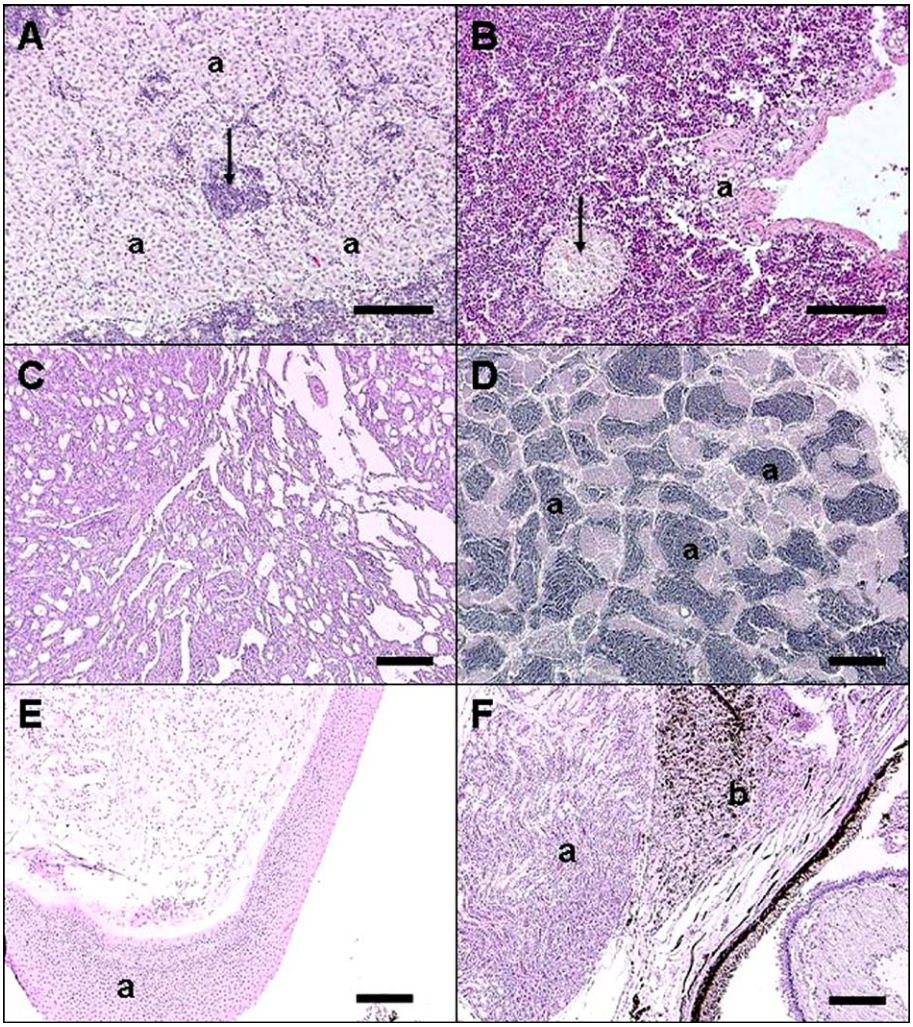

Fig. 5. Selected histological observations in fishes from the CRB. (A) Large area of hyperplastic interrenal (a) tissue which appears to engulf hematopoetic tissue (arrow) within the anterior kidney of a largemouth bass from Station 41. Scale bar = 100 μm. (B) Normal anterior kidney sections of bass often have little (a) or no observable interrenal tissue; however, macrophage aggregates (arrow) are observable. Scale bar = 100 μm. (C) Abnormal (neoplastic) testicular tissue in a carp from Station 503. Scale bar = 200 μm. (D) Normal testicular tissue from carp collected at Station 503, illustrating spermatozoa (a) as well as lighter-staining, less mature developmental stages. Scale bar = 200 μm. (E and F) Microscopic appearance of abnormal eyes in largescale sucker from Station 98. (E) Cornea (a) was thickened and inflamed. (F) Chronic inflammation of the choroid (a) and disrupted retina (b) was evident. Scale bars = 200 μm. All sections were stained with H&E.

0.10% at Station 96 to 0.25% at Station 42, and most individual bass (95%) had SSI values between 0.06% and 0.34%. Splenosomatic index in carp differed significantly among stations ($F_{8,\ 140}=6.85$, $P<0.01$) and between genders ($F_{1,\ 140}=30.1$, $P<0.01$). Mean SSI values ranged from 0.15% at Station 42 to 0.39% at Station 45 in female carp and 0.22% at Station 41 to 0.47% at Station 45 in male carp. The SSI values were > 0.60% in male carp from Stations 42, 45, 501, and 502, and SSI values were uniformly > 0.30% in male carp from Stations 45 and 502. In largescale sucker, SSI values differed significantly among stations ($F_{8,\ 120}=12.02$, $P<0.01$) but not between genders ($F_{1,\ 120}=0.00$, $P>0.05$). Station means ranged from 0.17% at Station 501 to 0.37% at Station 117, and most individuals (91%) had SSI values between 0.12 and 0.43%. Spleens in largescale sucker from Stations 98, 117, and 504 in the upper CRB were generally larger than those from other sites.

3.3.3. Macrophage aggregates

Splenic MAs were quantified in bass, carp, and largescale sucker. The effect of age on MA parameters was significant in bass ($P<0.05$); therefore, MA parameters were age-adjusted in bass only. All three

Table 5
Age-adjusted station means for splenic macrophage aggregate (MA) parameters in bass, carp, and largescale sucker

Taxon and station	MA-# (no./mm^2)		MA-A (μm^2)		MA-% (%)	
Bass						
117 - Creston, MT	4.84	ab	2374	ab	0.97	ab
41 - Hagerman, ID	7.86	cd	3239	bcd	2.30	cde
43 - Riggins, ID	10.41	d	4404	d	4.45	d
42 - Lewiston, ID	8.11	cd	4206	d	2.97	de
96 - Ice Harbor Dam, WA	7.62	bcd	2368	abc	1.75	bcd
503 - Vernita Bridge, WA	7.85	abcd	2698	abcd	1.88	bcde
97 - Pasco, WA	5.00	ab	2194	a	0.63	a
44 - Granger, WA	6.64	abc	3350	bcd	1.85	cd
502 - Warrendale, OR	4.28	abc	4416	cd	1.67	abcde
45 - Oregon City, OR	5.02	ab	3221	bcd	1.46	bc
505 - Portland, OR	4.29	a	3255	abcd	1.21	abc
ANCOVA						
Model (*df*=11, 111)	4.50**		4.29**		6.99**	
Station (*df*=10, 111)	3.53**		2.42**		4.18**	
Age (*df*=1, 111)	19.99**		27.00**		38.70**	
Carp						
41 - Hagerman, ID	3.04	a	2096	a	0.53	a
42 - Lewiston, ID	10.24	d	3411	bc	3.22	de
96 - Ice Harbor Dam, WA	6.81	bc	4163	cd	2.73	cde
503 - Vernita Bridge, WA	6.68	bc	3444	bc	1.82	bc
97 - Pasco, WA	6.91	bc	3180	bc	2.02	bcd
44 - Granger, WA	7.85	c	5284	d	3.84	e
502 - Warrendale, OR	5.68	b	2993	abc	1.47	bcde
45 - Oregon City, OR	5.92	bc	3519	bc	1.81	bcd
501 - Beaver Army, OR	6.32	abc	2939	abcd	0.64	b
ANOVA (*df*=8, 144)	6.41**		3.33**		7.92**	
Largescale sucker						
117 - Creston, MT	8.69	c	2743	a	2.21	abc
504 - Northport, WA	7.44	b	3414	a	2.43	bc
98 - Grand Coulee, WA	5.38	a	3384	a	1.69	ab
43 - Riggins, ID	6.38	abc	2790	a	1.52	ab
46 - Cascade Locks, OR	8.24	c	3621	a	2.82	c
506 - Vancouver, WA	7.12	abc	2695	a	1.34	a
502 - Warrendale, OR	5.29	abc	4802	a	2.51	abc
45 - Oregon City, OR	4.90	ab	3429	a	1.34	ab
505 - Portland, OR	5.51	a	2864	a	1.47	a
501 - Beaver Army, OR	8.01	c	2734	a	1.79	abc
ANOVA (*df*=9, 147)	2.45*		1.12ns		1.98*	

Stations are ordered upstream to downstream. Means were age-adjusted in bass only after ANOVA modeling determined age to be a significant factor. Shown are arithmetic mean MA density (MA-#) and geometric mean MA area (MA-A) and percent tissue occupied (MA-%) using analysis of variance (ANOVA) in carp and largescale sucker and analysis-of-covariance (ANCOVA) in bass (adjusted to the basin-wide mean age of 5.1 years). ANCOVA and ANOVA *F*-values and degrees-of-freedom (*df*) for the analyses (**, $P \leq 0.01$; *, $0.01 < P \leq 0.05$; ns, $P > 0.05$) are also presented. Means followed by the same letter within each column are not significantly different ($P > 0.05$, Fisher's protected LSD).

MA parameters differed significantly ($P < 0.05$) among stations for bass and carp, but only MA-# and MA-% were significant in largescale sucker (Table 5). Age-adjusted MA parameters were uniformly greater in bass from Stations 41, 42, and 43 in the Snake River Basin. All mean MA parameters were significantly lower in carp from Station 41, and carp from Station 44 had the greatest mean MA-A and MA-% (Table 5). Significant station differences in MA-# and MA-% were not consistent in largescale sucker; mean MA-# was greatest at Stations 46, 117, and 501, but mean MA-% was greatest at Station 46.

3.4. Reproductive biomarkers

3.4.1. Gonadal histopathology

Ovary samples from 74 female bass representing 11 stations were examined; gonadal stages 0–3 were present. Most fish (55%) were stage 1 and were from Stations 41, 42, 43, 97, 502, 503, and 505. Female bass from Stations 44, 45, 96, and 117 were more advanced (stages 2 and 3) than those from other sites (mostly stages 0 and 1). Ovary samples from 70 female carp from eight stations were examined; gonadal stages 0–3 were present. Most female carp (83%) were stage 2. Stage-0 and -1 female carp were present at Stations 41, 44, and 503, and one stage -3 fish was present at Station 502. Station 41 was the only site at which carp were predominantly less advanced (stages 0 and 1), but this was not related to collection date (Table 1). Ovary samples of 88 female largescale sucker from ten stations were examined; gonadal stages 1 (24%), 2 (50%), 3 (25%), and 5 (1%) were present. Stage-2 fish were predominant at Stations 45, 98, 117, 502, and 505, but most fish from Stations 501 and 504 were stage 3.

Female bass, carp, and largescale sucker had varying degrees of oocyte atresia. Atresia was typically <10% in bass, <20% in carp, and <8% in largescale sucker (Fig. 6). An ANOVA model containing the factors station, age, gonadal stage, and their interactions was significant only in carp. Although the model explained 57% of the total variation ($F_{20,\ 36}=2.36$, $P<0.05$), oocyte atresia in carp did not differ significantly among stations ($F_{1,\ 36}=0.32$, $P>0.05$).

Testes samples representing 58 male bass from 12 stations were examined; gonadal stages 0–3 were present. Most (93%) were in more advanced stages (stages 2 and 3) than females from the same site. Male bass from Stations 42, 43, and 503 were less advanced (stages 0 and 1) than those from other stations (all stage-2). Five male bass (all smallmouth) representing two stations had evidence of ovotestis as identified by the presence of developing oocytes in otherwise normal male testes. This intersex condition was detected in three of seven fish from Station 42 and in two of three

from Station 502. The intersex fish were in gonadal stages 0–3. Testes samples from 85 male carp from nine stations were examined; gonadal stages 2 (40%) and 3 (60%) were present. Like male bass, gonadal stages in male carp were generally more advanced than female carp from the same site. Testes samples from 69 largescale sucker from eight stations were examined, and gonadal stages 0 (3%), 2 (3%), and 3 (94%) were present. Stage-0 and -1 largescale sucker were identified at Stations 43 and 117, respectively, and reflected earlier collection dates at these sites (Table 1). Stage-3 largescale sucker were predominant at most stations including Stations 501 and 506, where fish were collected in spring. Ovotestis was not detected in male carp or largescale sucker.

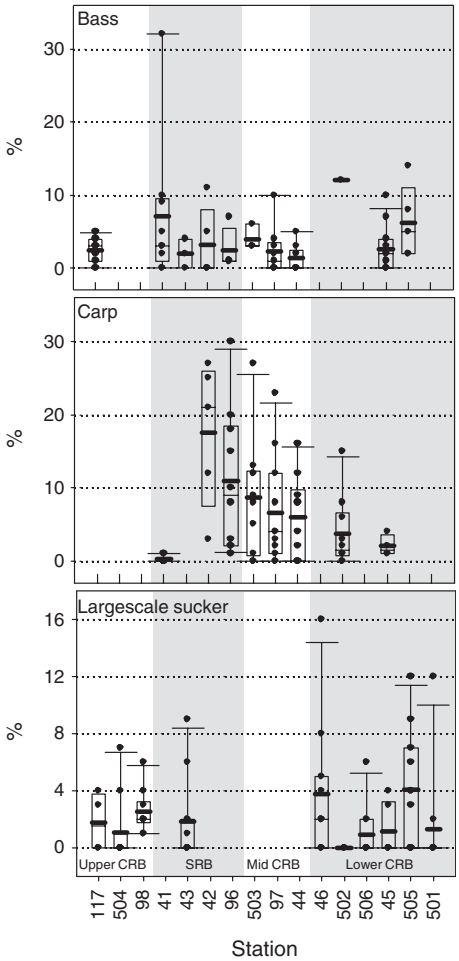

Fig. 6. Percentage of atretic oocytes in female bass, carp, and largescale sucker, by station. Shown for each group are points representing individual fish and the mean (heavy horizontal line), median (light horizontal line), interquartile range (box), and the 10th and 90th percentiles (whiskers). Stations are ordered from upstream to downstream and are grouped by sub-basin.

3.4.2. Gonadosomatic index (GSI)

The GSI can provide information about gonadal health and can be influenced by gender and gonadal stage. ANOVA models containing the factors station, gender, gonadal stage, and their interactions were significant for GSI in bass ($F_{38,\ 92}=16.08$, $P<0.01$), carp ($F_{27,\ 127}=12.95$, $P<0.01$), and largescale sucker ($F_{28,\ 127}=12.12$, $P<0.01$). The model explained 87% of the total variation in bass. The GSI did not differ among stations ($F_{8,\ 92}=0.77$, $P>0.05$) or between genders ($F_{1,\ 92}=0.85$, $P>0.05$), but was influenced by gonadal stage ($F_{1,\ 92}=16.74$, $P<0.01$). The GSI values in female bass ranged from 0.2% in a stage-1 fish from Station 41 to 7.2% in a stage-3 fish from Station 45 (Fig. 7). Ovaries were proportionately larger in female bass from Station 45 (>4%) and smaller in those from Station 42 (<1%) compared to other stations (Fig. 7). The GSI values were lower in male than female bass; individual GSI values in male bass ranged from 0.3% at Station 97 to 1.7% at Station 96 (Fig. 7). Unlike in female bass, GSI values did not appear to be correlated with gonadal stage in male bass.

Carp had proportionately larger gonads than bass, and carp ovaries were larger than male testes. The ANOVA model accounted for 73% of the total GSI variation in carp; GSI values differed significantly among stations ($F_{7,\ 127}=2.41$, $P<0.05$), but not between genders ($F_{1,\ 127}=2.75$, $P>0.05$). GSI values ranged from 0.6% at Station 41 to 20.5% at Station 502 in female carp and from 0.01% at Station 42 to 11.0% at Station 44 in male carp (Fig. 7). Male carp from Stations 44, 45, 501, and 502 were predominantly stage 3 and had greater GSI values; male carp from the other stations were predominantly stage 2. In general, GSI values increased upstream to downstream in male and female carp (Fig. 7); GSI values and gonadal stages were similar in male carp from Station 501, which were collected in April, to those from other stations.

Unlike bass and carp, GSI values were similar in female and male largescale sucker. The ANOVA model explained 73% of the total GSI variation in largescale sucker. The GSI values differed significantly among stations ($F_{9,\ 127}=5.26$, $P<0.01$) and gonadal stages ($F_{1,\ 127}=14.05$, $P<0.01$). GSI values in female largescale sucker ranged from 0.3% in a stage -1 fish from Station 43 to 13.4% in a stage-3 fish from Station 506 (Fig. 7). The relatively high GSI values in female fish from Stations 501 and 506 likely reflect the spring collection date, but GSI values in male largescale sucker were similar among stations regardless of collection date (Fig. 7; Table 1). GSI values in males

ranged from 0.8% in a stage-0 fish from Station 117 to 8.3% in a stage-3 fish from Station 501 (Fig. 7). Most largescale suckers were in stages 2 and 3 and had GSI values >2.9%; one stage-0 male had a GSI value of 0.77%.

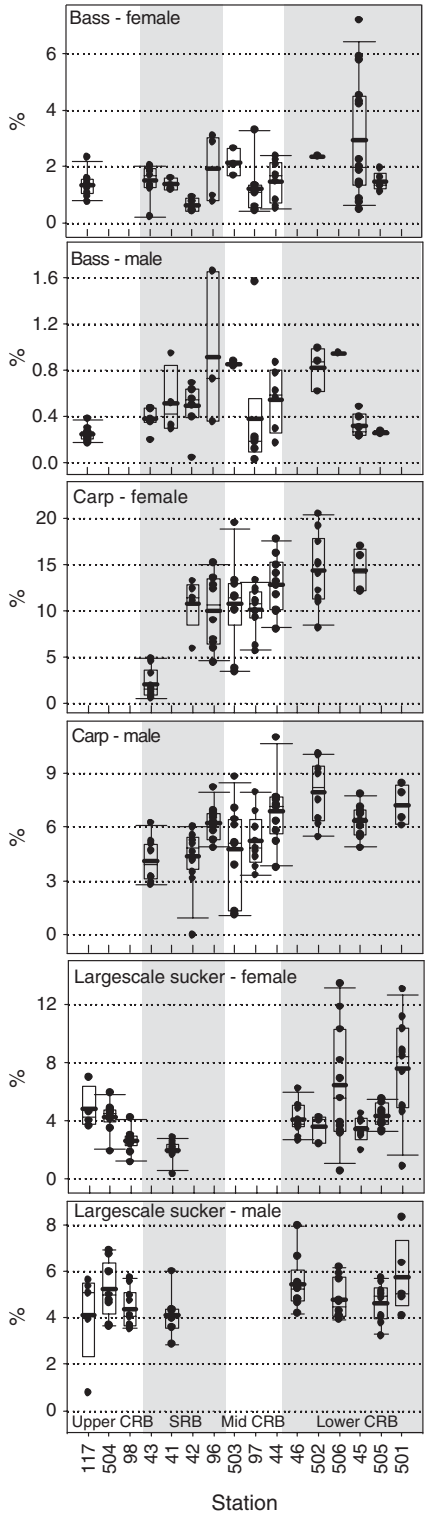

Fig. 7. Gonadosomatic index (GSI) values of female and male bass, carp, and largescale sucker, by station. Shown for each group are points representing individual fish and the mean (heavy horizontal line), median (light horizontal line), interquartile range (box), and the 10th and 90th percentiles (whiskers). Stations are ordered from upstream to downstream and are grouped by sub-basin.

3.4.3. Vitellogenin (vtg)

Concentrations of vtg >LOD in male bass (0.002 mg/mL), carp (0.005 mg/mL), and largescale sucker (0.0005 mg/mL) were rare; therefore, the data were not analyzed statistically. In addition, plasma samples were not available for fish from Stations 501 and 506. Concentrations in male bass ranged from <0.002 to 0.67 mg/mL and exceeded the LOD in at least one fish from Stations 42, 43, 96, 97, and 117 (data not shown). The maximum concentration was measured in a stage-2 fish from Station 97; all other concentrations were <0.1 mg/mL. Concentrations in male carp ranged from <0.005 to 0.04 mg/mL and exceeded the LOD in at least one fish from Stations 45, 502, and 503 (data not shown). The maximum concentration was measured in a stage-3 fish from Station 503; most concentrations >LOD were measured in stage-3 fish. Concentrations in male largescale sucker ranged from <0.0005 to 0.31 mg/mL and were >LOD in at least one fish from Stations 46, 98, 117, and 505 (data not shown). The maximum concentration was measured in a stage-3 fish from Station 98.

In female bass, carp, and largescale sucker, ANOVA models that included the factors station, gonadal stage, age, and their interactions were significant for vtg. The model explained 85% of the total vtg variation in bass ($F_{30,\ 62}=6.00$, $P<0.01$), 81% in carp ($F_{20,\ 55}=7.28$, $P<0.01$), and 61% in largescale sucker ($F_{25,\ 64}=2.39$, $P<0.01$). Station differences were not significant in any taxon, but gonadal stage was a significant factor in bass ($F_{1,\ 62}=4.52$, $P<0.05$). Concentrations in female bass ranged from <0.002 to 33.7 mg/mL (Fig. 8). Concentrations were uniformly >10 mg/mL in female bass from Station 503. Concentrations of vtg were generally greater in stage-2 and -3 bass than in stage-0 and -1 bass. Concentrations in female carp ranged from <0.005 to 7.4 mg/mL (Fig. 8); concentrations were ≥3.0 mg/mL in four stage-2 female carp from Stations 42 and 96. Vitellogenin concentrations in female largescale sucker ranged from 0.1 to 166 mg/mL (Fig. 8). Female largescale sucker from Stations 98, 502, and 505 had

concentrations <40 mg/mL and were predominantly in stages 1 and 2.

4. Discussion

4.1. Exposure indicators

Concentrations of most elemental contaminants, including As, Cd, and Zn, were relatively low. In contrast, concentrations of Hg, Se, and Pb exceeded toxicity thresholds at one or more sites (Table 6); elevated concentrations of these contaminants have been previously reported in the CRB (Walsh et al., 1977; Serdar et al., 1994; Schmitt et al., 1999; USEPA, 2002a).

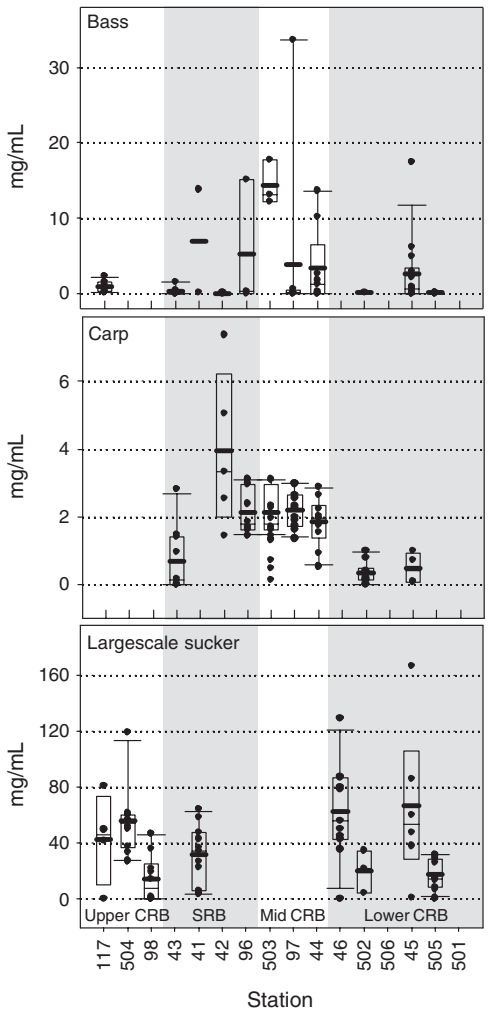

Fig. 8. Plasma vitellogenin (vtg) concentrations in female bass, carp, and largescale sucker, by station. Shown for each group are points representing individual fish and the mean (heavy horizontal line), median (light horizontal line), interquartile range (box), and the 10th and 90th percentiles (whiskers). Stations are ordered from upstream to downstream and are grouped by sub-basin.

Methylmercury, which is the most toxic form, represents most (>90%) of the Hg that occurs in fish (Wiener et al., 2002). Concentrations of total Hg were elevated (0.3–0.6 μg/g) in northern pikeminnow samples from Stations 43, 44, 502, and 503; historical NCBP concentrations in northern pikeminnow were <0.3 μg/g at Stations 43 and 44 but >0.3 μg/g at Stations 45 and 46 (Lowe et al., 1985). Our concentrations of total Hg in bass (0.06–0.44 μg/g) were relatively high but were similar to those in bass (0.22–0.36 μg/g) from a concurrent CRB study (USEPA, 2002a). Concentrations in largescale sucker (0.08–0.20 μg/g) were comparable to those measured in previous CRB investigations (Walsh et al., 1977; Serdar et al., 1994; USEPA, 2002a). Consumption advisories have been issued for fish from the Willamette River, Snake River, Brownlee Reservoir on the Snake River, and Lake Roosevelt on the CR as a result of elevated Hg concentrations in multiple species (Bastasch et al., 2002; USEPA, 2002c). Overall, Hg concentrations in bass and carp from the CRB were similar to those reported for the MRB in 1995 (Schmitt et al., 2002a) and the RGB in 1997 (Schmitt et al., 2005). Concentrations of Hg >0.3 μg/g in at least one sample from Stations 43, 44, 117, 501, and 502 may represent a threat to piscivorous birds (Barr, 1986), and concentrations were >0.1 μg/g in at least one sample from all stations, which may be a threat to piscivorous mammals (Yeardley et al., 1998).

Fish in embryonic and larval stages are at greatest risk from environmental Hg, partially due to maternal transfer (Wiener and Spry, 1996). Behavioral effects in fish with whole body Hg concentrations as low as 0.7 μg/g have been documented in laboratory studies (Kania and O'Hara, 1974). Other studies reported behavioral effects in fish with 5–10 μg/g of total Hg (Wiener and Spry, 1996; Wiener et al., 2002). Juvenile grayling (*Thymallus thymallus*) had permanent impairment of their feeding efficiency and competitive ability at whole body concentrations of 0.27 μg/g (Fjeld et al., 1998). Drevnick and Sandheinrich (2003) reported that female fathead minnows (*Pimephalas promelas*) given dietary methylmercury concentrations of 0.87 μg/g dw (0.22 μg/g ww assuming 75% moisture) led to a 10–fold increase in whole body Hg concentrations, which suppressed hormone concentrations and inhibited gonadal development. Many factors contribute to the uncertainty in estimating toxicity thresholds from tissue concentrations (Wiener et al., 2002), and further investigations would be needed to document such effects in CRB fish.

Table 6
Summary of chemical and biological findings indicative of exposure to contaminants by sub-basin and station (designations are relative)

Sub-basin and station	Contaminants and EROD activity	Fish health indicators	Reproductive biomarkers
Upper Columbia River Basin			
117 [a]	Hg (b), EROD (b,s)	SSI (s)	(None observed)
504 [a, b]	Hg (s), Pb (s), TCDD (s), EROD (fs-)	SSI (s), EL (s)	(None observed)
98 [a, b]	Hg (s), Pb (s), EROD (s)	SSI (s)	Vtg (ms)
Snake River Basin			
41 [c]	Hg (b)	EL (b,c), HAI (b)	(None observed)
43 [a]	Hg (b), Se (b), EROD (b,s)	(None observed)	(None observed)
42 [c]	Hg (b,c), DDE (c), chlordane (c), PCB (b,c)	SSI (fc-)	Ovt (mb)
96 [c]	Hg (b), DDE (c), TCDD (b,c), EROD (c)	(None observed)	Vtg (mb)
Middle Columbia River Basin			
503 [c]	Hg (b), Se (b), DDE (b,c), PCB (b,c), TCDD (b,c), EROD (c)	EL (c), HSI (fb)	Vg (mc)
97 [c]	Hg (b), Se (b,c), TCDD (b,c)	(None observed)	(None observed)
44 [c]	Hg (b), DDE (c,b), PCB (b)	EL (b,c), HAI (b)	(None observed)
Lower Columbia River Basin			
46 [a, b]	Hg (s), PCB (s), TCDD (s), EROD (s)	(None observed)	(None observed)
502 [d]	Hg (b,c), PCB (b,c), TCDD (c), EROD (c,fs)	EL (b,c)	Vtg (mc), ovt (mb)
506 [a, e]	Hg (s), PCB (s), TCDD (s)	(None observed)	(None observed)
45 [d]	Hg (b,c,fs), TCDD (b,c), EROD (b,c,fs)	EL (c), HAI (b)	(None observed)
505 [a]	Hg (b,s), TCDD (b,s), EROD (b,s)	EL (s)	(None observed)
501 [b, f]	DDE (mc,s)	EL (c)	(None observed)

Male (m) and female (f) bass, carp, and largescale sucker were collected from all sites unless otherwise indicated. If gender is not specified, then the indicated condition was present in both. Additional abbreviations: DDE, *p,p′*-DDE; chlordane, sum of *cis*- and *trans*-chlordanes and nonachlors; oxychlordane; and heptachlor epoxide; PCB, total PCBs; TCDD, dioxin-like activity as determined by H4IIE bioassay; Hg, total mercury; Pb, lead; Se, selenium; EROD, hepatic ethoxyresorufin *O*-deethylase activity; EL, external lesions; vtg, vitellogenin; HAI, health assessment index; SSI, splenosomatic index; ovt, ovotestis; MA, macrophage aggregates (one or more parameters); b, bass (*Micropterus* sp.); c, carp (*Cyprinus carpio*); s, largescale sucker (*Catostomus macrocheilus*); –, indicates that the response or condition was smaller or lower than most; all others larger or greater.

[a] No carp collected.
[b] No bass collected.
[c] No largescale sucker collected.
[d] No male largescale sucker collected.
[e] No female bass collected.
[f] No female carp collected.

Concentrations of Se were comparatively high (>0.6 μg/g) in fish from agricultural areas (Stations 43, 97, and 503) and confirmed previous CRB findings (Table 6). At Station 97, concentrations in carp (>0.9 μg/g) were consistent with historical NCBP concentrations from 1980–1986 (Schmitt et al., 1999). Concentrations in largescale sucker and northern pikeminnow in the lower CRB (<0.4 μg/g) were also similar to those reported previously (Walsh et al., 1977; USEPA, 2002a). Concentrations in bass were elevated (>0.9 μg/g) and comparable to those (0.48–0.71 μg/g) from a USEPA study in the CRB (USEPA, 2002a). Concentrations of Se in CRB fish were much lower than those (3–5 μg/g) reported for an agricultural site in the MRB (Schmitt et al., 2002a) and from the central parts of the RGB (Schmitt et al., 2005).

Diet is the primary route of Se exposure and toxicity in aquatic vertebrates (Lemly, 2002; Hamilton, 2004). Whole body concentrations of 8–16 μg/g dw (2–4 μg/g ww assuming 75% moisture) have be associated with reproductive failure in fathead minnows (Schultz and Hermanutz, 1990) and bluegill (*Lepomis macrochirus*; Gillespie and Baumann, 1986; Hermanutz et al., 1992; Coyle et al., 1993), and Se exposure in the egg stage or at hatch can affect larval survival. In addition, Se toxicity has been correlated with various conditions including swelling of gill lamellae, elevated numbers of lymphocytes, anemia, corneal cataracts, exopthlamus, pathological alterations of liver, kidney, heart, and ovary, reproductive failure, and teratogenic deformities of spine, head, mouth, and fins (Lemly, 2002; Hamilton, 2004). Toxicity thresholds associated with Se tissue concentrations are relatively low because of Se's high

toxicity and potential to bioaccumulate. Whole body concentrations of Se should not exceed 4 μg/g dw (1.0 μg/g ww assuming 75% moisture) to avoid toxicity to larval fish and 3 μg/g dw (0.75 μg/g ww assuming 75% moisture) to avoid toxicity to piscivorous wildlife (Lemly, 1996; 2002). Smallmouth bass from Stations 43 and 503 and carp and largemouth bass from Station 97 exceeded one or both of these thresholds.

Concentrations of Pb in largescale sucker from Stations 98 and 504 were elevated (>4 μg/g; Table 6). Station 504 at Northport, WA is located 31–33 km downstream from a smelting complex in British Columbia that historically discharged slag and slurry effluent containing elemental contaminants into the CRB (Bortelson et al., 1994). Lead concentrations in largescale sucker in this part of the CR are among the greatest reported in the US (Lowe et al., 1985; Schmitt et al., 1999, 2002b). Schmitt et al. (2002b) reported Pb concentrations of 1.34 μg/g in largescale sucker collected previously from Station 504, and Serdar et al. (1994) measured concentrations of 3.0–12.0 μg/g in largescale sucker collected near Station 504. Elevated concentrations in the northern hog sucker (*Hypentelium nigricans*; 4.57 μg/g) and black redhorse (*Moxostoma duquesnii;* 11.22 μg/g) collected near Pb smelters and mines in Missouri are comparable to those in largescale sucker from Station 504 (Schmitt et al., 1993). Concentrations in samples from other CRB sites were low (<0.1 μg/g) and consistent with previous studies (Walsh et al., 1977; Schmitt et al., 1999; USEPA, 2002a). Holcombe et al. (1976) determined that whole body Pb concentrations of 0.4 μg/g reduced hatchability and 4.0–8.8 μg/g reduced growth at various life stages in third generation brook trout (*Salvelinus fontinalis*). Largescale sucker from Stations 98 and 504 exceed these toxicity thresholds for Pb. Concentrations of other elemental contaminants including As, Cd, and Zn associated with mining in the CRB were comparatively low although Pb mine tailings containing those and other elemental contaminants have contaminated water, indigenous fish, and multiple bird species in the Coeur d'Alene and the Clark Fork Rivers in the upper CRB (Farag et al., 1994, 1995; Henny et al., 1994, 2000).

Residues from the pesticides DDT (as *p,p′*-DDE), chlordane, and dieldrin were the only organochlorine compounds regularly detected (>10%) in our samples. Relatively high concentrations of these residues were present in fish from agricultural areas of the lower Snake River and the middle CRB; the greatest concentrations were measured in carp from Station 42 (Snake River at Lewiston, ID) and in carp and bass from Station 44 (Yakima River at Granger, WA; Table 6).

DDT-derived residues (mostly as *p,p′*-DDE) were detected at all sites except Station 117. Concentrations were elevated (>0.8 μg/g) in fish from Stations 44 and 503 in the middle CR. However, comparison of 1997 concentrations with historical data revealed downward trends for *p,p′*-DDE at all sites, including Stations 44 and 97 (Schmitt et al., 1999). The higher concentrations at Stations 44 and 97 are most likely associated with historical DDT application in agricultural areas (Johnson et al., 1988a) as supported by a concurrent CRB study (USEPA, 2002a). Little or no *p,p′*-DDT was detected at these sites, indicating the continued weathering of residual DDT rather than the input of new material. In the lower CRB, concentrations of *p,p′*-DDE in fish (<0.01–0.68 μg/g) were greater than those previously reported (USEPA, 1992; Curtis et al., 1993; ODEQ, 1994). Concentrations of *p,p′*-DDE in carp and bass from the CRB were greater than those in the MRB (Schmitt, 2002b) and RGB (Schmitt et al., 2005) except for MRB stations in cotton-farming regions of the lower Mississippi River valley. Concentrations of *p,p′*-DDE exceeded 0.15 μg/g, a concentration potentially toxic to the most sensitive avian species (Anderson et al., 1975), in one or more samples from all CRB sites except Stations 98, 117, and 504. Concentrations of 1–3 μg/g are potentially hazardous to most avian wildlife (Blus, 1996) and were detected in bass from Station 44 and carp from Station 503. Wildlife criteria for total DDT as low as 0.20 μg/g have been suggested (Newell et al., 1987), and toxic effects have been reported at concentrations as low as 0.5 μg/g in several fish species (see review by Jarvinen and Ankley, 1999).

Comparatively low concentrations of *o,p′*-DDT (0.01–0.043 μg/g) were detected in samples from Stations 503, 505, and 506. Largescale sucker from Station 506 also contained traces (0.01 μg/g) of *o,p′*-DDE and *o,p′*-DDD. Like many other pesticides and their metabolites, *o,p′*-DDD is weakly estrogenic even though the *o,p′*-homologs have historically been considered relatively benign (Guillette et al., 1996; Toppari et al., 1996; Tyler et al., 1998; Ackerman et al., 2002). The cumulative risk to fish and wildlife represented by concentrations of *o,p′*-DDT is unknown.

Total chlordane concentrations were relatively low (<0.13 μg/g) and confirmed results from previous investigations (USEPA, 1992; Munn and Gruber, 1997). Nevertheless, the maximum concentration in male carp from Station 42 slightly exceeded the toxicity threshold (0.3 μg/g) to protect predatory fish and piscivorous birds (Eisler, 1990). Dieldrin concentrations were >0.02 μg/g in carp from Stations 42 and 96 in the lower Snake River and in carp and bass from Station 44

in the Yakima River. Similar concentrations were reported in largescale sucker and carp from the middle CR (Munn and Gruber, 1997) and the lower CR (USEPA, 1992; Curtis et al., 1993; ODEQ, 1994). These chlordane concentrations do not represent a significant threat to either fish or piscivorous wildlife based on available criteria (Peakall, 1996; Jarvinen and Ankley, 1999).

Concentrations of PCBs were relatively high (>0.5 μg/g) in bass from Stations 42, 44, and 505, northern pikeminnow from Stations 502 and 503, and largescale sucker from Station 506 (Table 6). Similar concentrations (0.03–1.3 μg/g) were reported by other CRB studies (Tetra Tech, 1996; Clark and Maret, 1998; Foster et al., 2001). Concentrations were significantly less than historical concentrations at all NCBP stations (Schmitt et al., 1999), although concentrations in smallmouth bass (<0.03–0.64 μg/g) and largescale sucker (<0.03–0.75 μg/g) were greater than those reported in a contemporaneous study (USEPA, 2002a). The New York State Department of Environmental Conservation (NYSDEC) wildlife guideline for total PCBs in fish is 0.11 μg/g (Newell et al., 1987), a concentration exceeded by at least one sample from all stations except Stations 43 and 97. All concentrations were less than those known to cause adverse effects on reproduction and development in fish (5 μg/g; Monosson, 2000). Mink (*Mustela vison*) fed Great Lakes fish or fish products with PCB concentrations of 0.48 μg/g had inferior reproductive performance and offspring survival (Hornshaw et al., 1983). Concentrations at some sites exceeded one or more of these toxicity thresholds even though concentrations of PCBs appear to be decreasing.

Concentrations of TCDD-EQs were generally low (<5 pg/g) in the upper CRB and upper Snake River Basin. However, a dioxin consumption advisory in Lake Roosevelt for lake whitefish (*Coregonus clupeaformis*) has been in place since 1994 (USEPA, 2002c). TCDD-EQs were not strongly correlated with PCB concentrations, which indicates the presence of one or more other dioxin-like compounds in the samples. TCDD-EQs >5 pg/g may be hazardous to piscivorous wildlife based on the potential for reproductive impairment and biomagnification (Nosek et al., 1992; Tillitt et al., 1996); TCDD-EQs >30 pg/g may be toxic to fish (Walker et al., 1996; Whyte et al., 2004). Most TCDD-EQs in our study were similar to those reported in fish from reference sites (Giesy et al., 1995; van den Heuvel et al., 1995; Schmitt et al., 2002a) and lower CR sites (Bonn, 1998). The technical product of the herbicide Dacthal® contains 2,3,7,8-TCDD and hexachlorobenzene (HCB) as impurities (Cox, 1991). The greatest TCDD-EQ (43 pg/g) was measured in a carp sample from Station 96, where high concentrations of Dacthal® have been reported (Schmitt et al., 1999). Station 96 was also the only station where HCB was detected (>0.01 μg/g; data not shown). Concentrations of TCDD-EQ and HCB may therefore reflect the historical use of Dacthal® near this site. Overall, piscivorous wildlife may be at risk to TCDD and similar compounds in the lower CRB.

We considered the normal ranges of hepatic EROD activity to be 0–16 pmol/min/mg in female bass, 0–22 pmol/min/mg in male bass, 0–4 pmol/min/mg in female carp, and 0–6 pmol/min/mg in male carp based on data from the MRB (Schmitt et al., 2002a) and 0–15 pmol/min/mg in female and male largescale sucker based on a review by Whyte et al. (2000). EROD activity in at least one fish from all stations was greater than these criteria and indicates exposure to AhR agonists. Mean EROD activity in bass exceeded these criteria in one or both genders from Stations 43, 45, 117, and 505, as did at least one bass from all sites except Stations 41 and 503 (Table 6). Mean EROD activity in carp exceeded the criteria in one or both genders from Stations 45, 96, 502, and 503, as did at least one carp from all sites except Station 97 (Table 6). Mean EROD activity in largescale sucker exceeded the previously cited criteria at all sites (Table 6). EROD activities were not correlated with PCBs or TCDD-EQs for bass, carp, or largescale sucker, which indicates that fish from most CRB stations had been exposed to polycyclic aromatic hydrocarbons or other AhR agonists.

4.2. Fish health indicators

The quantitative fish health indicators used in this study have been widely used and discussed in the literature. Most CRB bass and carp had CF values between 1.0 and 2.0, which are typical values for these taxa (Carlander, 1969, 1977; Blazer et al., 2002); CF values in largescale sucker were similar (0.9–1.2) and are also typical for other sucker species (Carlander, 1969). The HSI values in bass ranged from 0.53% to 3.17% and were >2.0% in fish from Station 503 (Table 6). The liver normally constitutes 1–2% of the body in most fish (Gingerich, 1982). The HSI values in bass from Station 503 (CR at Vernita Bridge, WA) were the largest we have measured; HSI values >2.0% were rare in bass from the MRB (Blazer et al., 2002) or RGB (Schmitt et al., 2005). Enlarged livers have been reported in various fish species after exposure to contaminants including PCBs, PAHs, and both bleached and unbleached kraft

mill effluents (Adams and McLean, 1985; Schmitt and Dethloff, 2000; Sepúlveda et al., 2001, 2003). PCBs, TCDD-EQ, and EROD activity were also elevated in fish from Station 503 (Table 6).

In bass, SSI values of 0.05–0.2% were typical, but values >0.4% were documented in bass from Stations 42, 44, and 505. The SSI values were low (<0.2%) in carp from Station 42, and individual carp with relatively large spleens were found at Stations 45, 501, and 502. Splenosomatic index values were high (>0.4%) in largescale sucker from the upper CRB relative to other sites, and many largescale sucker from Stations 98 and 504 had external lesions that were similar to those characteristic of infections. The co-occurrence of lesions and enlarged spleens has been documented by Goede and Barton (1990). Conversely, exposure to PCBs, PAHs, and metals has been associated with contracted (i.e.; smaller) spleens (Blazer et al., 2002).

Reference information on external lesions and MA parameters in freshwater fishes is limited. In addition, and as noted by Schmitt et al. (2005), external lesion or abnormality data vary among studies depending on both field personnel expertise (Leonard and Orth, 1986) and differing anomalies characterized and recorded (Karr, 1981; Fournie et al., 1996; Sanders et al., 1999). Our criteria are comparable to the MRB (Blazer et al., 2002) and RGB (Schmitt et al., 2005) studies, and are similar to the widely used deformities, erosion, lesions, and tumors (DELT) component of the Index of Biotic Integrity (IBI; Karr, 1981; Sanders et al., 1999). In general, we noted sites at which >85% of the bass, carp, or largescale sucker had grossly visible lesions (Table 6). Lesion frequencies in CRB fish were greater than those in fish from both the MRB (Blazer et al., 2002) and RGB (Schmitt et al., 2005).

The HAI in carp and bass have been used in previous studies (Adams et al., 1993; Coughlan et al., 1996; Blazer et al., 2002; Schmitt et al., 2005), but it has only been used once in largescale sucker. HAI scores in individual largescale sucker ranged from 0 to 60 in a previous fish health study in the lower CRB (Tetra Tech, 1996). Based on these studies and our data, we assumed that sites with mean HAI scores ≤20 were un-impacted or minimally impacted sites, sites with means >50 were intermediate, and sites with means >70 were heavily impacted. The mean HAI score was >70 in bass from Stations 41, 44, and 45 but were <70 in carp and largescale sucker from all sites. However, HAI scores >70 were documented in individual bass, carp, and largescale sucker from most sites. Overall, the HAI scores and frequencies of external lesions indicate some degradation of fish health at many CRB sites.

Fournie et al. (2001) suggested a value of >40 splenic MA/mm^2 in at least one fish from a site as a threshold indicative of possible effects due to hypoxia or sediment contamination in marine and estuarine fishes. This value was used as a benchmark for carp and bass in the MRB, with recognition of the fact that threshold effects may differ in freshwater species (Blazer et al., 2002), and that it does not consider the importance of a fish with a few large aggregates (Schmitt et al., 2005). Nevertheless, and consistent with findings from the MRB (Blazer et al., 2002) and RGB (Schmitt et al., 2005), no CRB fish contained >40 splenic MA/mm^2 (MA-#), and station means for all species were <11 MA/mm^2. Mean MA-A (2096–5294 μm^2) and MA-% (0.6–4.4%) in CRB bass and carp were also similar to previous investigations (Blazer et al., 2002; Schmitt et al., 2005). Increases in MA parameters have been associated with contaminants in laboratory and field studies (see review by Wolke, 1992; Blazer et al., 1994; 1997), but can vary with fish size and age, nutritional status (Wolke et al., 1985), age (Brown and George, 1985; Blazer et al., 1987; Couillard and Hodson, 1996), and exposure time (many studies as reported in Schmitt and Dethloff, 2000).

Few incidences of confirmed tumors or other grossly visible indications of exposure to toxic chemicals were found in CRB fish. Papillomas (benign tumors of the skin) were noted in five largescale sucker collected near Northport, WA (Station 504). Greater prevalence of papillomas in various fish species have been reported in populations exposed to industrial and sewage effluents (Kortet et al., 2002). The poorer health (e.g., papillomas, enlarged spleens) of largescale sucker from Station 504 may be related to effluents from a smelting complex and a pulp mill in British Columbia, but more detailed investigations would be required to identify causation in such situations.

4.3. Reproductive biomarkers

Reproductive biomarker measurements were compared to those in the MRB (McDonald et al., 2002) and RGB (Schmitt et al., 2005); controlling for gonadal stage differences is important when making such comparisons (McDonald et al., 2002). Fish were collected in fall 1997 except at Stations 501 and 506, where they were collected in spring 1998. Carp and largescale sucker typically spawn during May and June when the water reaches appropriate temperatures. Female largescale sucker from Stations 501 and 506 had greater GSI values than those from other stations, but most were only early to mid-vitellogenic (stages 2

and 3). We expected more advanced gonadal stages in fish from Stations 501 and 506 because the fish would have spawned soon after the collection date. Male carp and largescale sucker from Station 501 and 506 did not have greater GSI values compared to fish collected in the fall but did contain mature spermatozoa (stage 3), indicating that they were reproductively active even though they were in the same gonadal stages as fish collected in the fall. Altered water temperatures associated with dams, urbanization, logging, and agriculture may affect the spawning cycle of fish in the CRB (USEPA, 2001). Nevertheless, more information is needed to understand the annual cycle of gonadal development in carp and largescale sucker in the CRB.

The GSI differed in bass, carp, and largescale sucker; gonads constituted a substantially greater proportion of the total body mass in carp and largescale sucker than in bass. As noted, GSI values were greater in female largescale sucker from Stations 501 and 506, where they were collected in spring 1998, than those from other sites. After accounting for stage differences, other GSI differences were only evident in a few fish of all taxa. Other studies have reported that changes in the GSI occur after exposure to a variety of endocrine-modulating substances and may be more apparent as the fish approach spawning season (Sepúlveda et al., 2001; Orlando et al., 2004).

Although oocyte atresia is a normal physiological event in all fish, it can become a pathological condition following exposure to certain environmental contaminants (Johnson et al., 1988b; Kirubagaran and Joy, 1988; Cross and Hose, 1988, 1989). McDonald et al. (2002) defined atresia ≥25% for female carp and >10% for female bass as high. Atresia was generally <10% in bass, <20% in carp, and <8% in largescale sucker from the CRB. High individual values were measured in bass (>10%) from Stations 41, 42, 502, and 505, in carp (>20%) from Stations 42, 96, 97, and 503, and in largescale sucker (>12%) from Stations 46, 501, and 505.

To the best of our knowledge, the background occurrence of intersex male bass has not been established. We documented where intersex fish were found and at what frequency, although there are no criteria for comparison. Five male smallmouth bass from two sites (Stations 42 and 502) were identified as histologically abnormal and having ovotestis. Ovotestis were reported in largemouth and smallmouth bass from the MRB (McDonald et al., 2002) and the Hudson River (Baldigo et al., 2000) and in largemouth bass from the RGB (Schmitt et al., 2005). We did not detect ovotestis in male carp from any CRB station, which is consistent with findings in the MRB (McDonald et al., 2002) and RGB (Schmitt et al., 2005).

Vitellogenin is an important yolk protein for developing embryos. Plasma vtg concentrations in female carp and bass were not abnormally low at any CRB station. Concentrations in female largescale sucker >20 mg/mL occurred frequently, and mean concentrations were >50 mg/mL in females from Stations 45, 46, and 504. These are high relative to the other taxa, but typical concentrations are unknown for this species. However, these values were greater than mean concentrations in female longnose sucker from the CRB (<17 mg/mL; Hinck et al., 2004a) and the Yukon River Basin (<4.9 mg/mL; Hinck et al., 2004b). Causes for differences in vtg concentrations among species are unknown; more information is required to determine if vtg concentrations in female largescale sucker were within normal ranges or were the result of exogenous estrogens.

Elevated plasma vtg concentrations in male fish may indicate exposure to xenoestrogens (Denslow et al., 1999). Concentrations of vtg in multiple individual male bass from Station 96, male carp from Stations 502 and 503, and male largescale sucker from Station 98 exceeded 0.01 mg/mL. These concentrations may represent estrogenic responses in these fish resulting from exposure to environmental conditions (Table 6). Concentrations of vtg in all intersex male bass were ≤0.01 mg/mL. In male fish, vtg concentrations in the range of early to mid-vitellogenic females (>0.8 mg/mL) were considered to be high in previous studies (McDonald et al., 2002; Schmitt et al., 2005). Similarly, vtg concentrations in male bass from Station 97 (0.7 mg/mL) and male largescale sucker from Stations 98 (0.5 mg/mL) and 117 (0.3 mg/mL) were abnormally high.

5. Conclusions

Overall, we saw little evidence that CRB fish had been exposed to extremely high concentrations of toxic chemicals, which is consistent with findings from the MRB (Schmitt, 2002a) and RGB (Schmitt et al., 2005). Fish from some CRB sites may have responded to chronic contaminant exposure as indicated by fish health indicator and reproductive biomarker results. Previous studies have concluded that fish and wildlife may be at risk from organic (e.g., *p,p′*-DDT, chlorinated dioxins, PCBs) and inorganic (e.g., Hg, As) contaminants from urban runoff and industrial discharges in the lower CRB (Rosetta and Borys, 1996; Tetra Tech Inc., 1996; Wentz et al., 1998; Lower Columbia River Estuary Partnership, 1991). Our findings support these

conclusions. Concentrations of Hg, PCBs, and TCDD-EQs in fish may represent a hazard to fish and piscivorous wildlife in the lower CR and Willamette River; EROD activities were also elevated in fish from many of these same sites and further indicate that PCBs and dioxins are a concern in the lower CRB.

Agricultural practices have also degraded water quality by increasing nutrient loads, pesticide concentrations, or both in the Willamette River (Wentz et al., 1998) and Yakima River Basins (Rinella et al., 1993, 1999; Ebbert and Embrey, 2002). As a result, water quality impairments for *p,p′*-DDT, *p,p′*-DDE, and dieldrin have been reported for parts of the Yakima River and its reservoirs that are in predominantly agricultural areas; PCB and Hg impairments have also been reported (USEPA, 2002b). A consumption advisory has been issued for *p,p′*-DDT and *p,p′*-DDE in all bottom fish from the Yakima River, all its tributaries, and agricultural drains between the city of Yakima, WA and its confluence with the CR (USEPA, 2002c). Concentrations of *p,p′*-DDE, dieldrin, and PCBs in fish from the Yakima River near Granger, WA were elevated in our study and are consistent with other findings.

A smelting complex in Trail, British Columbia has discharged slag and slurry effluent into the CR, resulting in heavy metal contamination as far downstream as Northport, WA and Grand Coulee Dam (Bortelson et al., 1994; Schmitt et al., 2002b). Also in the upper CRB, tailings and other mining wastes containing As, Cd, Cu, Pb, and Zn have contaminated the Clark Fork-Pend Oreille and Spokane River basins (Maret and Skinner, 2000; Beckwith, 2002), and fish kills in the Clark Fork since 1984 have been attributed to the toxic effects of these trace metals (Maret and Skinner, 2000). Concentrations of PCBs are a concern in fish tissues from the Spokane River and exceed the protective criterion for piscivorous wildlife (MacCoy, 2001). Mercury impairments have been previously reported from the international border to Grand Coulee Dam and have resulted in consumption advisories for walleye in Lake Roosevelt (USEPA, 2002c). A consumption advisory has also been issued for dioxins in lake whitefish from Lake Roosevelt (USEPA, 2002c); the probable source of dioxin is a pulp mill in British Columbia (Schneider, 2002). Concentrations of Hg and dioxin-like activity as measured by TCDD-EQs in largescale sucker from Station 504 (Northport, WA), located upstream from Lake Roosevelt, were also elevated and support other findings in this area. Previous investigations have reported that Hg and pesticides may be hazardous to fish and wildlife in the Snake River (Clark et al., 1998; USEPA, 2002a); our findings also support these conclusions. Moreover, concentrations of Hg and *p,p′*-DDE have not decreased significantly in fish from Station 44 (Granger, WA) compared to historical concentrations.

The human population in the Pacific Northwest continues to grow rapidly. This growth will likely expand the magnitude and type of chemical contaminants released in the CRB even though our data indicate declining concentrations of certain persistent contaminants at some sites. Water quality in the lower CR will likely continue to decline as industrial and municipal discharges increase, as reflected in the numerous impairments listed for the region. Toxic trace elements may leach into river systems as a result of mining operations and irrigated agriculture. Concentrations of contemporary-use pesticides including atrazine, azinphos-methyl, carbaryl, and diazinon in some agricultural areas already exceed chronic toxicity guidelines for the protection of aquatic wildlife (Ebbert and Embrey, 2002). Biological responses to these and other chemicals may occur at concentrations less than current standards, and cumulative effects are largely unknown (e.g., McDonald et al., 2000; Scholz et al., 2000). Our results indicate that the biological endpoints we measured were consistent with chronic exposure to chemicals at some CRB sites. These biological responses would be expected to increase in magnitude as concentrations of pesticides and other substances increase, which may ultimately place fish populations at risk. Results from this study and other investigations indicate that continued monitoring is needed to identify consistently degraded sites and those with emerging problems, such as polybrominated diphenyl ethers (PBDEs; Johnson and Olson, 2001; Ikonomou et al., 2002), in the CRB. Focused investigations are also needed to document chemical sources, interactions with other factors, and cause–effect relations. The results from this study should be examined together with other investigations to identify potential underlying relations between contaminant exposure and responses.

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