Reproductive and Morphological Condition of Wild Mink (Mustela vison) and River Otters (Lutra canadensis) in Relation to Chlorinated Hydrocarbon Contamination

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We assessed chlorinated hydrocarbon contamination of mink and river otters on the Columbia and Fraser River systems of northwestern North America, in relation to morphological measures of condition. We obtained carcasses of mink and river otters from commercial trappers during the winters 1994–1995 and 1995–1996. Necropsies included evaluation of the following biological parameters: sex, body mass and length, age, thymus, heart, liver, lung, spleen, pancreas, kidney, gonad, omentum, adrenal gland and baculum masses, baculum length, and stomach contents. Livers were analyzed, individually or in pools, for residues of organochlorine (OC) pesticides, polychlorinated biphenyls (PCBs), dibenz-p-dioxins, and dibenzofurans. Contaminant levels were relatively low compared to those documented in other North American populations, although they ranged higher than those detected during an earlier survey (1990–1992) of these regional populations. Body condition varied slightly among collection regions, but showed no relationship with contaminant burden. Mink from the upper Fraser River had less fat stores and also had some of the lowest OC contamination levels observed. Similarly, a few individuals with enlarged livers and kidneys had low contaminant levels. Although a few individual animals with gross abnormalities of reproductive systems did not show high levels of contamination, there was a significant negative correlation between total PCB concentrations (as Aroclor 1260) and baculum length in juvenile mink ($r = 0.707; p = 0.0335; n = 8$). The association of juvenile baculum length with eventual reproductive success is unknown, but further characterization of reproductive organ morphology and relationship to contaminants should be undertaken in a larger subset of these populations.

The sensitivity of mink (Mustela vison) to polychlorinated biphenyls (PCBs) was identified in the early 1970s when reproductive failure of ranch mink was attributed to a diet of contaminated Great Lakes fish (1–3). Further studies in which adult ranch mink were fed PCB-contaminated fish from Saginaw Bay, Lake Huron, showed a dose–response relationship between PCB concentration in food and reproductive failures, measured as reduced litter sizes, increased kit mortality, and reduced average kit weight (4). Declines of European otter (Lutra lutra) populations (5–7) and North American mink populations (8,9) have been tentatively linked to elevated environmental PCB concentrations.

Recent laboratory studies have examined mechanisms by which aryl hydrocarbon receptor (AhR) agonists such as 2,3,7,8-TCDD (dioxin) can alter sexual maturation and reproduction in various mammalian species (10). Results include a variety of feminizing effects in male offspring and less pronounced effects on female reproductive systems (11). Those studies suggest that, for example, feminizing effects of TCDD and other compounds may be discernible in field-exposed individuals.

The principal objective of this study was to evaluate possible relationships between tissue levels of contaminants and measurements of certain organs, particularly reproductive organs. Given the presence of several organochlorine (OC) pesticides, polychlorinated dibenzo-p-dioxins (PCDDs) and dibenzofurans (PCDFs), and PCBs in pooled liver samples of mink and river otters (Lutra canadensis) previously collected by Elliott et al. (12), those species were again collected from trappers during the 1994–1995 and 1995–1996 winter seasons along the reaches of both the Fraser and Columbia river systems in British Columbia. Collections were also extended to the Kootenay River, a tributary of the Columbia, and the lower Fraser River. Hepatic chlorinated hydrocarbon concentrations were assessed, along with several biological parameters. Biological characterization included sex, age, body mass and length, organ condition indices, baculum size in males, and stomach content analysis. Results from analysis of the same animals for heavy metals are being published elsewhere (13).

Materials and Methods

Sample collection. Skinned mink and river otter carcasses were collected from commercial trappers during the winters of 1994–1995 (November–March) and 1995–1996 (November–January). Traps lines were located on the Fraser River near Prince George and Chilliwack, British Columbia, Canada; on the Kootenay River near Creston and Cranbrook, British Columbia, on the Slocan River (a tributary to the Kootenay River); and on the Columbia River between Revelstoke and Trail, British Columbia. In the Fraser Valley, a suburban/rural environment near the terminus of the Fraser River, mink and otters were obtained from licensed trappers without commercial traps lines, and who were permitted to remove nuisance animals. Specific collection locales are indicated in Figure 1. Over the two seasons, a total of 26 mink and 32 river otters were collected on the three river systems. Six of the mink were not subjected to the same chemical and biological measurements described below, but are included in the data sets regarding sex and age ratios.

Trappers were provided with collection kits to ensure consistent and adequate quality of tissue storage. Collection kits consisted of aluminum foil, garbage bags, labels for recording the date and location of collection and the sex of the trapped specimen, and detailed instructions on collection handling and storage. Skinned carcasses were frozen before shipment by air to the laboratory.

Necropsies. All mink and otters collected in the winter of 1995–1996, as well as otters collected the previous winter on the Kootenay River, were thawed and a gross
necropsy performed by a veterinarian (C.R.S.). The carcasses (without pelts) were sexed and weighed, and total length (nose to tip of tail) and tail length were measured. After external examination for gross abnormalities (including a count of toes), each carcass was dissected to assess organ condition and internal structure. One upper and one lower canine tooth were removed for dental cementum analysis of age (14.15); unfortunately, some of the otter teeth were damaged during extraction, so that ages were not obtained for all animals. Thymus, heart, lung, liver, spleen, pancreas, gonads, kidney, omentum, and adrenal glands were excited with hexane-washed knife and forceps, weighed, and examined for gross pathology. Livers were placed in hexane-washed jars and frozen for chemical analysis as described below. Stomach contents were assessed on a superficial level (e.g., if fish scales were encountered, they were not identified to genus or species level). In males, the baculum was excised, cleaned with a digestive enzyme and also by hand to remove all adhering tissues, and then air-dried for several months. Cleaned and dried baculae were weighed (mass) with an electronic balance and measured (length) with an electronic micrometer. Some animals were received with heads missing, so that there was some variability in sample size for the numerous data sets.

**Analysis of hepatic organochlorine contamination.** Excised livers from mink and river otters were analyzed at the Pacific Environmental Science Centre, Environment Canada (North Vancouver, BC) for chlorinated organic compounds. The organic contaminants included the pesticides aldrin, dieldrin, endosulfan, endrin, heptachlor, heptachlor epoxide, p,p'-DDD, p,p'-DDE, p,p'-DDT, and PCBs. Livers were homogenized, acetone-extracted, and partitioned with 2% NaCl. Extracts were then treated, as necessary, with florisil, concentrated sulfuric acid, activated copper, and/or mercury to remove interferences. Glass columns prepared with glass wool, 2% deactivated florisil, and heat-treated sodium sulfate were washed with 50 ml hexane. A 2-ml sample extract was loaded, and the first and second fractions were eluted with 35 ml hexane and 150 ml 1:1 hexane/dichloromethane (DCM), respectively. The first fraction contained PCBs, p,p'-DDE, aldrin, and heptachlor, and the second fraction contained the remainder of the pesticides. Fractions were roto-evaporated, and then quantitated using a HP-5890 series II high resolution gas chromatograph with electron capture detection (Hewlett-Packard, Cupertino, CA). Method blanks were included with each run, and 20% of the samples were split and analyzed as duplicates. Discrepancies of greater than 20% among duplicates were not acceptable. External 0.1- and 1.0-μg/g PCB standards were analyzed twice during each sample run. Instrument calibration was completed at three concentrations using pesticide mixture and Aroclor 1260 standards. Average recoveries were 89%, with coefficients of variation of 1.4-12.7%. Minimum detection limits were 0.01 μg/g for PCBs and 0.002 μg/g for pesticides. All concentrations discussed in the text were expressed on a wet weight basis. Otters' livers were also pooled by region and analyzed at Zenon Analytical (Burnaby, BC) using high-resolution gas chromatography/mass spectrometry (GC/MS) for PCDDs, PCDFs, and co-planar PCBs (16). Those data are included in the Appendix.

**Statistical evaluation.** Biological data collected from necropsies of mink and river otters were grouped by collection region and river to permit statistical interpretation. In all cases, tests for normality and homogeneity of variances were completed, and transformations were deemed unnecessary. Organ condition or somatic indices were calculated for kidney, liver, and spleen by dividing the organ mass by a corrected total body mass
and multiplying by 100. The corrected body mass was derived by subtracting the kidney, liver, and spleen masses from the total skinned carcass mass. Organ somatic indices were compared within sexes among regions using analyses of variance (ANOVA). Organ masses were also compared using the more conservative analyses of covariance (ANCOVA), with total body mass as the covariate. Where a significant difference in organ mass or somatic index was seen among regions, an unplanned multiple comparison using Tukey’s honestly significant difference test (17) was conducted. Gross body measures (length and mass) were also submitted to ANOVA, grouped according to sex and region, but without consideration of age differences. When significant differences among regions were found, an ANCOVA was conducted with age as the covariate. Analyses of covariance were also conducted on adrenal, omentum, testes, and baculum masses and baculum length, with total body mass as the covariate. All tests were performed with SYSTAT 5.0 (18).

We tested several biological parameters for associations with each other and with contaminant body burdens using Pearson correlation matrices. Correlations were assessed for significance using a Bonferroni adjustment when more than two associations were being evaluated simultaneously. Occasionally, an association between just two variables was retested separately, in which case a Bonferroni adjustment of the probability value was not necessary.

**Results**

Twenty-six mink and 32 river otters were collected over the course of the two winter seasons. Sex ratios for the captured mink and otters were dominated by males; 73% of mink and 59% of otters collected were males (Table 1). In addition, of those individuals whose ages could be determined, the majority captured had been born within the year (i.e., juveniles). In the subsample of animals with determined ages, 74% of mink and 42% of otters were juveniles. Sample sizes were too small to compare sex or age ratios among study areas.

**Hepatic chlorinated hydrocarbon contamination.** Most mink livers, which were all collected along the Fraser River, contained detectable concentrations of PCBs and DDT-related compounds (r-DDT, primarily p,p’-DDT), and several also contained dieldrin (Table 2). Most otter livers from the Kootenay, Columbia, and lower Fraser Rivers contained detectable concentrations of PCBs, p,p’-DDT, and heptachlor epoxide (Table 3). None of the otters from the upper Fraser had detectable p,p’-DDT and only one had detectable PCBs; four animals had detectable heptachlor epoxide concentrations.

In both mink and otters, hepatic concentrations of OC pesticides and PCBs were correlated in males (mink: $r = 0.685$, $p = 0.002$; otters: $r = 0.876$, $p < 0.001$), but not in females ($p > 0.05$).

A subset of the animals collected was also analyzed for PCDDs, PCDFs, and non-ortho PCB congeners; those results are compiled in the Appendix along with some previously published data. Concentrations of 2,3,7,8-TCDD and TCDF were low (<5 pg/g) in all samples. The pattern of higher chlorinated PCDDs and PCDFs varied among sites, with the pools from the lower Fraser valley having much higher relative concentrations of octaCDD and 1,2,3,4,6,7,8-hexaCDD and higher chlorinated PCDFs than other sites.

**Morphological characterisation.** Reproductive tract abnormalities were seen in a few

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**Table 1.** Mean body weights and lengths (mean ± standard error) for mink and otters trapped on the Fraser, Columbia, and Kootenay Rivers of British Columbia, 1994–1996

| Species   | N  | Percent of total | Mean weight (kg) | Mean length (cm) |
|-----------|----|------------------|------------------|------------------|
| Mink      |    |                  |                  |                  |
| Males     | 19 | 73               | 0.88 ± 0.07      | 56.5 ± 1.2       |
| Females   | 7  | 27               | 0.90 ± 0.04      | 49.2 ± 1.0       |
| Juveniles | 14 | 54               | 0.74 ± 0.10      | 53.9 ± 1.1       |
| Yearlings | 3  | 11               | 1.00 ± 0.13      | 60.8 ± 3.9       |
| Adults    | 2  | 8                | 0.64 ± 0.05      | 49.8 ± 0.25      |
| Otter     |    |                  |                  |                  |
| Males     | 19 | 59               | 7.19 ± 0.41      | 107.5 ± 2.4      |
| Females   | 13 | 41               | 6.83 ± 0.47      | 107.4 ± 2.08     |
| Juveniles | 7  | 22               | 6.01 ± 0.66      | 103.8 ± 3.1      |
| Yearlings | 5  | 16               | 7.04 ± 0.55      | 114.0 ± 2.6      |
| Adults    | 5  | 16               | 7.19 ± 0.48      | 112.1 ± 2.1      |

*Seven mink (27%) and 15 otters (46%) were of unknown age.

**Table 2.** Mean and range of chlorinated hydrocarbon contaminants (in μg/g wet weight) in livers from mink collected along the Fraser River in British Columbia, 1994–1996

| Sample              | PCBs (Aroclor 1260) | Dieldrin | r-DDT |
|---------------------|---------------------|----------|-------|
| Downriver Fraser (n=8) | 0.07 ± 0.03 | 0.01 ± 0.01 | 0.07 ± 0.10 |
| (0.03–0.12)         | (<0.002–0.27)       | (0.01–0.30) |
| Upper Fraser (n=12)  | 0.08 ± 0.13 | 0.01 ± 0.01 | 0.03 ± 0.05 |
| (0.01–0.46)         | (<0.002–0.048)      | (0.002–0.18) |

**Abbreviations:** PCBs, polychlorinated biphenyls; r-DDT, p,p’-DDT-related compounds, including p,p’-DDT, DDD, and DDE. No detections are expressed as detection limit (0.002 for pesticides, 0.01 for PCBs). Aldrin, endosulfan, endrin, heptachlor, and p,p’-DDE were not detected in any samples.

**Table 3.** Mean and range of organochlorine contaminants (in μg/g wet weight) in livers from otters collected along the Columbia and Kootenay Rivers in British Columbia

| Sample              | Percent lipid | PCBs | Heptachlor epoxide | r-DDT |
|---------------------|---------------|------|-------------------|-------|
| Kootenay (n=12)     | 0.9           | 0.15 ± 0.18 | 0.01 ± 0.03 | 0.12 ± 0.33 |
| (<0.01–0.61)        | (<0.002–0.11) | (<0.002–1.69) |
| Lower Columbia (n=1)| 1.2           | 1.44  | <0.002            | <0.002 |
| Upper Columbia (n=4)| 0.9           | 0.12 ± 0.10 | 0.01 ± 0.01 | 0.04 ± 0.03 |
| (<0.01–0.21)        | (<0.002–0.01) | (0.02–0.08) |
| Lower Fraser (n=3)  | 0.5           | 0.14 ± 0.19 | 0.01 ± 0.02 | 0.01 ± 0.04 |
| (<0.01–0.35)        | (0.003–0.007) | (0.006–0.01) |
| Upper Fraser (n=6)  | 0.7           | 0.03 ± 0.07 | 0.01 ± 0.01 | <0.002 |
| (<0.01–0.17)        | (<0.002–0.004) |
| Kootenay 1990–1991  | 3.0           | 0.064b | <0.001            | <0.001 |
| Columbia 1990–1991  | 2.1–2.8       | 0.12–0.58b | <0.001 | 0.002–0.004 |
| Upper Fraser 1990–1991 | 3.4        | 0.038b | <0.001            | <0.001 |

**Abbreviations:** PCBs, polychlorinated biphenyls; r-DDT, p,p’-DDT-related compounds, including p,p’-DDT, DDD, and DDE. No detections are expressed as detection limit (0.002 for pesticides, 0.01 for PCBs). Aldrin, endosulfan, endrin, heptachlor, and p,p’-DDE were not detected in any samples.

*Otter samples collected from the Kootenay, Columbia, and upper Fraser Rivers in 1990–1991 (12).

Analyses of PCBs as Aroclor 1260 were not conducted; value or range represents sum-PCB congeners (see Elliott et al. (12)).

Environmental Health Perspectives • Volume 107, Number 2, February 1999
male mink and otters. In one mink and one otter, a testicle had not descended into the scrotum, but was instead located in the inguinal canal; the otter was also missing a kidney on the same side as the undescended testis. In two mink, no testes or bacula were found. Another otter was missing one testicle and had an underdeveloped second testicle. No abnormalities were seen in the reproductive tracts of the females. Of the two female mink and four female otters known to be of breeding age, only one otter was pregnant. She carried two fetuses measuring 6 cm from crown to rump and weighing 9.9 and 10.5 g. The corpora lutea were not counted in other females, so it is not known whether they had undergone copulation and successful ovulation that year.

Three mink showed other lesions not associated with reproductive organs. One male mink had a healed umbilical infection with some associated abdominal adhesions. Another mink had a thickened, scarred area on the mucosa of its stomach, and a third had an enlarged, hemorrhagic mesenteric lymph node.

There were no significant differences in average testes mass, baculum length, or baculum mass in male mink or river otters from the various collection regions (Table 4). There appeared to be an association between baculum size and age (juveniles vs. yearlings) in both species, but correlations were only significant in mink (mass and age: \( r = 0.805, p = 0.024 \); length and age: \( r = 0.774, p = 0.047 \)). There were several strong correlations among the various reproductive organ sizes. Male mink average testes mass was significantly correlated with total body mass (juveniles: \( r = 0.987, p<0.001 \); all males: \( r = 0.813, p = 0.008 \), and baculum length and mass were correlated with each other (all males: \( r = 0.785, p = 0.015 \)). In male otters, total body mass, average testes mass, and baculum length were all significantly associated with each other; Pearson correlations ranged from 0.696 to 0.927, with associated Bonferroni-adjusted probabilities of 0.05–0.001. Baculum mass was associated with average testes mass \( (r = 0.927, p<0.001) \) and baculum length \( (r = 0.866, p = 0.001) \), but not total body mass.

The morphological condition of sampled animals showed few differences among collection regions. Although there was a substantial amount of variability in extent of subcutaneous and intraabdominal fat deposits among animals, none showed signs of severe emaciation, such as atrophy of fat. Seven of the nine mink collected from the upper Fraser River had empty stomachs, were significantly lighter \( (p = 0.011, \text{males only}) \), and had significantly less omentum tissue \( (p = 0.027) \) than counterparts from the lower reaches of the river. However, signs of emaciation were not present. In addition, male otters from the lower Fraser had larger livers than otters from the upper Columbia River \( (p = 0.012) \), and male mink from the Kootenay had larger livers and kidneys than mink from the Fraser River \( (p = 0.008 \text{ and } p = 0.04, \text{respectively}) \). No mass differences in adrenal tissue were found in either species.

**Associations among biological parameters and contaminant burdens.** Few significant relationships among biological parameters and OC pesticide and PCB concentrations were found in mink, and none at all were found in otters. There were no differences in contaminant body burdens between males and females in either species \( (p=0.05) \). In both male and female mink, there was a significant correlation between age and total pesticide contamination. Juveniles of both sexes were much less contaminated with dieldrin and DDT-related compounds than yearling males \( (r = 0.788; p = 0.024) \) and adult females \( (r = 0.945; p = 0.046) \). There were also significant correlations between male mink baculum size and hepatic PCB concentration, and baculum mass and total pesticide concentration. However, baculum mass associations with contamination were a function of age; the four variables were highly intercorrelated \( (mass \text{ and age: } r = 0.805, p = 0.024; \text{mass and PCBs: } r = 0.781, p = 0.002; \text{mass and pesticides: } r = 0.778, p = 0.002; \text{age and pesticides, as shown above}) \), and baculum mass along with contaminant concentrations increased with increasing age. When juveniles alone were considered, baculum mass was not associated with either contaminant variable. Conversely, baculum length of juveniles \( (r = 0.707; p = 0.033) \) decreased with increasing PCB contamination \( \text{(Fig. 3)} \), and there was no association between PCBs and age. There were no other significant associations between hepatic OC pesticide or PCB concentrations and liver,

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**Figure 2.** TCDD toxic equivalents (TEQs) in livers of river otters collected from British Columbia, 1994–1996. Abbreviations: CDFs, chlorinated dibenzo-p-dioxins; CDDs, chlorinated dibenzofurans; PCBs, polychlorinated biphenyls; KOT, Kootenay River; L, lower; U, upper; COL, Columbia River; FV, Fraser Valley; a and b refer to pooled otter samples.

**Table 4.** Testes mass and baculum size (mean ± standard error) in male mink and river otters trapped in the Fraser, Columbia, and Kootenay Rivers in British Columbia, 1994–1996.

| Species | Age  | No | Testes mass (g) | Baculum length (mm) | Baculum mass (g) |
|---------|------|----|----------------|---------------------|------------------|
| Mink    | All  | 13 | 0.52 ± 0.14    | 40.1 ± 0.6          | 0.17 ± 0.02      |
|         | Juveniles | 9 | 0.39 ± 0.12    | 39.3 ± 0.7          | 0.15 ± 0.01      |
|         | Yearlings | 3 | 1.02 ± 0.40    | 42.7 ± 0.2          | 0.26 ± 0.04      |
| Otter   | All  | 17 | 5.0 ± 1.3      | 86.9 ± 3.1          | 4.1 ± 0.6        |
|         | Juveniles | 3 | 1.9 ± 0.5      | 81.3 ± 2.3          | 2.0 ± 0.02       |
|         | Yearlings | 3 | 8.4 ± 2.8      | 94.7 ± 2.1          | 6.6 ± 0.6        |
|         | Adults | 2  | ND             | 100.5 ± 0.5         | 6.5 ± 0.2        |

NO, not determined. Baculum size is reported for cleaned, air-dried specimens.
kidney, spleen, adrenal, omentum, or testes masses in mink, nor any organ size in otters. Most of the contaminant information was obtained on composite samples of otters, and direct comparisons with biological parameters cannot be made.

Discussion

Hepatic concentrations of chlorinated hydrocarbon contaminants in mink and otters collected in British Columbia ranged slightly higher than mean levels determined from the same regions during a 1990–1991 survey; however, they were about an order of magnitude less than samples from the lower reaches of the Columbia River (12) and from other North American populations (21). Although higher or equivalent to maximum means recorded previously for our study areas, international TEQ values of 27 pg/g wet weight in otter livers from the lower Fraser River and 20 pg/g in otter livers from the lower Columbia are below those considered to cause effects on reproduction of ranch mink. Leonards et al. (22) calculated critical body residues of 160–200 pg TEQs/g wet weight for litter size and kit survival effects in mink. The PCB concentration reported by Jensen et al. (23) to reduce reproductive success in female mink by 50% (50 mg/kg liver weight) was still twofold higher than the most contaminated female otter in our study (24 mg/kg liver weight).

The pattern of relatively elevated concentrations of octaCDD and other higher chlorinated PCDDs and PCDFs, particularly in the otter pools from the lower Fraser Valley, probably indicates the heavy use of chlorophenolic biocides by many years for the forest industry in that area. Eggs of ospreys (Pandion haliaetus) collected in other areas of the Fraser River basin had particularly high concentrations of octaCDD (up to 7,000 pg/g in a single egg) and other higher chlorinated PCDDs and PCDFs, which correlated significantly with pentachlorophenol concentrations (24).

Although we found some abnormalities in male reproductive systems, those individuals consistently had among the lowest concentrations of chlorinated hydrocarbons for both species. Where cryptorchidism (the incomplete descent of testicles) was observed, it was probably due to genetic defects sometimes associated with this condition in domestic animals (25). In some of the cases where testicles were missing and the condition of the carcass was too poor to identify a remnant of the vas deferens, the possibility of it having been removed during skinning cannot be excluded, although we considered this unlikely. However, where abnormalities were paired, as in two otters each with one undescended testicle and either a missing kidney or a missing second testicle, genetic explanations or skinning artefacts are even less likely. In particular, missing organs are more likely to have developed during fetal development. In such cases, we would not necessarily expect a relationship between presence of abnormalities and persistent contaminants in the body, as effects on reproductive development may be more related to female parental body burden.

Recent studies have reported that in utero exposure of rats to relatively low concentrations of TCDD and related PCBs can have significant effects on development of the reproductive system of offspring, without any subsequent lactational or feeding exposure (10,11). Effects on male offspring included reductions in testis weights and fertility, and effects on females included genital abnormalities, decreases in fecundity and litter size, and increases in pup mortality. If such effects are largely a function of maternal exposure, they would not be correlated necessarily with chlorinated hydrocarbon body burden in a wild mammal that had been feeding independently for at least a few months.

Nevertheless, Henny et al. (26) recently found significant correlations between PCBs and other OC contaminants and reproductive abnormalities such as smaller testes and bacula in river otters collected from the Columbia River in Oregon. Based on average hepatic PCB concentrations (as Aroclor 1260) for the Oregon otters (0.04–0.65 ppm wet weight), otters from British Columbia were within the same contaminant range for that particular PCB measure, yet they showed no association between baculum size and PCB contamination. Although it is difficult to compare biological parameters without knowing the natural regional and genetic variability, average baculum size of juvenile otters from British Columbia (8.13 cm; 2 g) was roughly equivalent to juvenile otters from the Columbia River (8.3 cm; 2.62 g) and much less than that of the juveniles that Henny et al. (26) collected from an Oregon reference population (9.52 cm; 5.82 g). It is apparent that further sampling of both exposed and reference populations is required to establish confidence in such findings for wild mustelids.

Even though contaminant body burdens were lower (on a wet weight basis), juvenile male mink from British Columbia displayed a similar correlation between hepatic PCB concentration (Aroclor 1260) and baculum length as Henny found in otters (26). This may indicate that mink are more sensitive than otters to PCB-induced reproductive effects in males, especially considering that, overall, mink had lower chlorinated hydrocarbon levels than otters. Alternatively, it may be that the (unknown) contaminant burden of the pregnant female is more important in determining baculum size of offspring. Past studies of OC effects on ranch mink have focused on female reproductive potential, commonly evaluating end points such as kit survival, weight of kits, number of kits whelped, adult female weight changes, and length of gestation period (27–30). Smits et al. (29) evaluated testicular development in young male mink exposed to bleached kraft mill effluent in the diet and found no effects in either male or female reproductive potential at the applied concentrations. On the whole, effects on male reproductive development and integrity have not been assessed, despite the evidence of significant effects in breeding females. The association found here in juvenile males exposed to relatively low concentrations of OCs suggests that reproduction may also be affected in males. Of course, whether baculum size has any bearing on reproductive success is unknown.

We recognize that the observed association between mink baculum size and hepatic PCB contamination is based on a relatively small sample size and that such a result may be a spurious finding given the large number of correlations that were studied. The observed correlation may also be a coincidental relationship, with an effect caused more by genetic and ecological variability. Nevertheless, our findings, together with the independent report of a similar association in otters from the lower Columbia (26), suggest a need for further investigation of possible effects of PCBs on reproductive development of male mustelids.

Several of the most contaminated individuals collected in our study were females, notably otters from the lower Fraser River and the lower Columbia River. From the perspective of identifying risks of reproductive
toxicity associated with chlorinated hydrocarbons in wild populations, those females are probably the most important subgroup to evaluate. Thus, biases inherent in sample collection via commercial traps may confound attempts to conservatively evaluate contaminant levels in wild mustelids.

The combined results of biological and chemical investigations presented here suggest that the mink and river otter populations using these major river systems in British Columbia do not appear physiologically or reproductively compromised because of chlorinated hydrocarbon accumulation. However, the relationship between PCBs and mink baculum size warrants further study. Also, even without postulating a link with OC contaminants, the occurrence of potentially serious developmental abnormalities in several systems (reproductive, digestive, and renal) in a relatively high proportion (about 10%) of individuals from wild populations of both species should be investigated. Finally, carcasses from commercial trapping are biased against sampling adult females, which is likely the critical component of the population impacted by contaminants. Alternative sampling methods should be considered to better survey adult and female segments of the populations.

REFERENCES AND NOTES

1. Harttous MG. Great Lakes fish now suspect as mink food. Am Fur Breeder 38:24 (1995).
2. Aulerich RJ, Ringer RK. Some effects of chlorinated hydrocarbon pesticides on mink. Am Fur Breeder 43(8):10-11 (1970).
3. Aulerich RJ, Ringer RK, Seagrín HL, Youatt WG. Effects of feeding coho salmon and other Great Lakes fish on mink reproduction. Can J Zool 49:811-816 (1971).
4. Heaton SN, Bursian SJ, Giese JP, Tillitt DE, Render JA, Jones PD, Verbrugge DA, Kubiat TJ, Aulerich RJ. Dietary exposure of mink to carp from Saganay Bay. I: Effects on reproduction and survival and potential risks to wild mink populations. Arch Environ Contam Toxicol 28:334-343 (1995).
5. Masaon CF. Water pollution and otter distribution: a review. Lutra 32:97-131 (1989).
6. Masaon CF, O'Sullivan WM. Organochlorine pesticide residues and PCBs in otters (Lutra lutra) from Ireland. Bull Environ Contam Toxicol 38:387-393 (1982).
7. Smit MD, Leonards PEG, van Ural B, de Jongh AWJ. PCBs in European Otter (Lutra lutra) Populations. Rpt no R-947. Amsterdam: Institute for Environmental Studies, Vrije Universiteit, 1994.
8. Henny CJ, Blouw LJ, Gregory SV, Stafford CJ. PCBs and organochlorine pesticides in wild mink and river otters from Oregon. In: Worldwide Furbearer Conference Proceedings (Chapman JA, Parsley D, eds). Baltimore, MD: University of Maryland Press, 1991:1763-1769.
9. Addison EM, Fox GA, Gilbertson M, eds. Proceedings of the Expert Consultation Meeting on Mink and Otter. Great Lakes Science Advisory Board's Ecological Committee Report to the International Joint Commission, 5-6 March 1991, Windsor, ON, Canada. Windsor, ON, Canada: International Joint Commission, 1991.
10. Peterson RE, Theobald RM, Kimmel GL. Development and reproductive toxicity of dioxins and related compounds: cross-species comparisons. CRC Crit Rev Toxicol 23:263-329 (1993).
11. Roman BL, Peterson RE. Developmental male reproductive toxicity of 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) and PCBs. In: Reproduction and Development Toxicology (Korach KS, ed). New York Marcel Dekker, 1998:503-624.
12. Elliott JE, Henny CJ, Harris ML, Wilson LK, Norstrom RJ. Chlorinated hydrocarbons in livers of American mink (Mustela vison) and river otter (Lutra canadensis) from the Columbia and Fraser River basins, 1990-92. Environ Monit Assess (in press).
13. Harding LE, Harris ML, Elliott JE. Heavy and trace metals in wild mink (Mustela vison) and river otter (Lutra canadensis) captured on rivers receiving metal discharges. Bull Environ Contam Toxicol 81:500-607 (1986).
14. Stephenson AB. Age determination and morphological variation of Ontario otters. Can J Zool 55:1577-1583 (1977).
15. Matson GM. Workbook for Cementum Analysis. Milltown, NJ: Matson's Laboratory, 1981.
16. U.S. EPA. Test Methods for Evaluating Solid Waste, Physical/Chemical Method. 3rd ed. EPA SW846.

Washington, DC:U.S. Environmental Protection Agency, 1988.
17. Sokal RR, Rohlf FJ. Biometry. The Principles and Practice of Statistics in Biological Research. New York:W. H. Freeman and Company, 1981.
18. Wilkinson L. SYSTAT: The System for Statistics. Evanston, IL:SYSTAT Inc., 1990.
19. Ahlberg UG, Becking GD, Birmbaum LS, Brouwer A, Derks HJ, Feeley M, Goral G, Hanberg A, Larsen JC, Lien AKO, et al. Toxic equivalence factors for dioxin-like PCBs. Chemosphere 286:1049-1087 (1994).
20. Sjö S. Polychlorinated biphenyls (PCBs), dibenzofurans and related compounds: environmental and mechanistic considerations which support the development of toxic equivalency factors (TEFs). Crit Rev Toxicol 21:51-88 (1990).
21. Halbrook RS, Woff LL, Hubert GF Jr, Ross S, Brasetel WE. Contaminant concentrations in Illinois mink and otter. Ecotoxicology 5:103-114 (1996).
22. Leonards PEG, de Vries TH, Minnaard W, Stuhl-land S, de Voogt P, Cofino WP, van Straalen NM, van Hattum B. Assessment of experimental data on PCB-induced reproduction inhibition in mink, based on an isomer- and congener-specific approach using 2,3,7,8-tetrachlorodibenzo-p-dioxin toxic equivalence. Environ Toxicol Chem 14(4):639-652 (1995).
23. Jensen S, Kihistrom L, Ohlson M, Lundberg C, Gerber J. The effects of PCB and DDT on mink (Mustela vison) during the reproductive season. Ambio 6:239 (1977).
24. Elliott JE, Machmer MM, Henny CJ, Wilson LK, Norstrom RJ. Contaminants in otters from the Pacific Northwest. I: Trends and patterns in polychlorinated dibenz-p-dioxins and dibenzofurans in eggs and plasma. Arch Environ Contam Toxicol 26:620-631 (1994).
25. Thomson RG. Special Veterinary Pathology. Toronto, Ontario:B.C. Decker Inc., 1988.
26. Henny CJ, Grove RA, Hedstrom OR. A Field Evaluation of Mink and River Otter on the Lower Columbia River and the Influence of Environmental Contaminants. Final Report to the Lower Columbia River Bi-State Water Quality Program (Portland, OR). Corvallis, OR:National Biological Service, Forest and Rangeland Ecosystem Science Center, 1996.
27. Hornshaw TC, Aulerich RJ, Johnson HE. Feeding Great Lakes fish to mink: effects on mink and accumulation and elimination of PCBs by mink. J Toxicol Environ Health 11:933-946 (1983).
28. Aulerich RJ, Bursian SJ, Napolitano AC. Biological effects of epidermal growth factor and 2,3,7,8-tetrachlorodibenzo-p-dioxin on developmental parameters of neonatal mink. Arch Environ Contam Toxicol 17:27-31 (1988).
### Chlorinated hydrocarbons and morphology of wild mustelids

| 2,3,7,8-TCDD | 1,2,3,6,7,8-HexaCDD | OctaCDD | Sum of other PCDDs | 2,3,7,8-TCDF | 2,3,4,7,8-PentaCDF | Sum of other PCDs |
|--------------|---------------------|---------|-------------------|-------------|-------------------|------------------|
| <3.0         | <7.0                | <30     | <8.4              | <2.6        | <3.0              | <7.7             |
| <1.90        | <4.0                | <13     | <4.8              | <1.9        | <1.7              | <4.0             |
| 0.39         | 2.5                 | 27      | 8.3               | 0.4         | <0.2              | 21.5             |
| 3.00         | 2.7                 | 13      | 7.7               | 1.2         | 5.6               | 27.6             |
| 0.41         | 5.7                 | 15      | 15.7              | <0.4        | 5.5               | 26.1             |
| 0.36         | 2.4                 | 23      | 12.0              | <0.3        | 2.7               | 17.0             |
| 0.96         | 57.0                | 160     | 132.3             | 0.5         | 15.0              | 115.9            |
| <0.19        | 4.1                 | 180     | 40.1              | <0.2        | 1.5               | 29.0             |
| <0.19        | 0.7                 | 5       | 2.8               | <0.2        | 0.3               | 1.2              |
| <2           | <4-<4               | <15-16  | <3-<23            | <3          | <3-<3              | <10              |
| <1.0         | <0.4-21.5           | 1.4-186.2| <2.7-124.5       | <3          | <0.2-3.1          | <2.0-16          |
| <2           | <4                  | <15     | <3                | <3          | <3                | <4               |
| 11           | 6                   | <15     | <3                | <3          | 19                | 2                |
| <1           | 11                  | 20      | 16                | <3          | <0.4              | 3.8              |

29. Smits JEG, Wobeser GA, Schiefer HB. Physiological, reproductive and pathological effects of dietary bleached pulp mill effluent on mink (Mustela vison). Environ Toxicol Chem 14(12):2095-2105 (1996).

30. Tillitt DE, Gale RW, Meadows JC, Zajicek JL, Peterman PH, Heaton SN, Jones PD, Bursian SJ, Kubiak TJ, Giesy JP, Aulerich RJ. Dietary exposure of mink to carp from Saginaw Bay. 3: Characterization of dietary exposure to planar halogenated hydrocarbons, dioxin equivalents, and biomagnification. Environ Sci Technol 30(1):283-291 (1996).

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