CONTAMINANT-RELATED DISRUPTION OF VITAMIN A DYNAMICS IN FREE-RANGING HARBOR SEAL (PHOCA VITULINA) PUPS FROM BRITISH COLUMBIA, CANADA, AND WASHINGTON STATE, USA

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(Received 22 November 1999; Accepted 10 April 2000)

Abstract—Marine mammals can bioaccumulate high concentrations of lipophilic environmental contaminants, such as polychlorinated biphenyls (PCBs), polychlorinated dibenzo-para-dioxins (PCDDs), and polychlorinated dibenzofurans (PCDFs), through the diet. Both laboratory and wildlife studies have shown that these persistent chemicals can disrupt the regulation of vitamin A (retinol), a dietary hormone required for immune function, reproduction, growth, and development. To determine whether environmental contaminants affect the circulatory vitamin A dynamics of free-ranging harbor seals (Phoca vitulina), we live-caught 61 pups from British Columbia, Canada, and Washington State, USA, and obtained blood and blubber biopsy samples. Harbor seal pups from Washington State were six times more contaminated with total PCBs than pups from British Columbia and had significantly lower circulatory retinol levels. However, when data were corrected for differences in nursing status and analyzed as ungrouped sets of data, circulatory retinol levels were positively correlated with contaminant levels in the blubber of nonnursing pups. This increase in retinol may have resulted from a mobilization of liver vitamin A stores into circulation following exposure to milk-derived contaminants; this has been observed in laboratory animals exposed experimentally. The contaminant-related disruption of vitamin A dynamics observed in our study occurs at a time when vitamin A is required for growth and development.

Keywords—Vitamin A Retinol Polychlorinated biphenyls Biomarker Phoca vitulina

INTRODUCTION

Organochlorines, including polychlorinated biphenyls (PCBs), polychlorinated dibenzo-para-dioxins (PCDDs), and polychlorinated dibenzofurans (PCDFs), are structurally similar groups of ubiquitous environmental pollutants. Top predators, including marine mammals, can bioaccumulate relatively high concentrations of these persistent chemicals in their blubber, sometimes leading to adverse effects such as reproductive impairment, endocrine disruption, developmental abnormalities, and immunotoxicity [1]. The latter is suspected to have played a role in recent virus-associated die-offs of marine mammal populations living in highly polluted areas [1]. While establishing cause-and-effect relationships represents a considerable challenge in studies of wildlife exposed to complex environmental mixtures of contaminants, biomarkers of exposure and effect can signal whether marine mammal populations are at risk. Vitamin A (retinol), a dietary hormone that is easily measured in blood samples, represents one such biomarker. Circulatory retinol levels in laboratory animals and wildlife have been shown to be adversely affected by both acute and chronic exposure to specific PCB congeners [2,3] and 2,3,7,8-tetrachlorodibenzo-para-dioxin [4–7] as well as by exposure to complex environmental mixtures of contaminants [8,9].

Vitamin A is a highly regulated micronutrient that is required for a wide variety of physiological functions, including growth and development [10]. In marine mammals, the majority of vitamin A is stored as retinyl esters in the liver and blubber [11,12], while lower concentrations are found in target tissues such as the pancreas, kidney, retina, lung, and spleen [13]. Vitamin A is delivered to these tissues by its transport protein, retinol binding protein (RBP), which circulates as a 1:1 molar complex with transthyretin (TTR), the transport protein for the thyroid hormone, thyroxine [14]. Together, TTR and RBP regulate the amount of vitamin A that is delivered to target tissues, maintaining circulatory vitamin A levels relatively constant over a wide range of dietary intakes and liver stores [15].

Despite the tightly controlled regulation of vitamin A, contaminant exposure can disrupt vitamin A dynamics by one of two elucidated mechanisms. Many PCBs, PCDDs, and PCDFs can bind and activate an intracellular cytosolic protein, called the aryl hydrocarbon receptor [16]. The resulting complex is translocated to the nucleus, where it binds to DNA and alters the transcription of specific proteins, some of which regulate vitamin A storage and catabolism [17]. In addition, some organochlorine hydroxy metabolites are structurally similar to thyroxine and have a high affinity for its binding site on TTR [18]. Hydroxy metabolites that displace thyroxine induce a conformational change in TTR, causing a dissociation of RBP-retinol [19], which is then more readily filtered by the kidney [15]. As a result of these two mechanisms, contaminant exposure can reduce liver vitamin A stores, disrupt the circulatory transport of retinol to target tissues, and increase the catabolic breakdown and excretion of vitamin A and its metabolites [2,4–7,19]. Although other mechanisms are likely involved, it is interesting to note that many of the characteristics of contaminant exposure resemble symptoms of vitamin A deficiency [7].

While the mechanisms of contaminant-related vitamin A toxicity have been elucidated in laboratory animals, a disrup-
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**MATERIALS AND METHODS**

**Sample collection**

Between 1996 and 1997, harbor seal (Phoca vitulina) pups from Johnstone Strait, the Strait of Georgia, and Queen Charlotte Strait, British Columbia, Canada (48°20′–50°50′N, 123°20′–127°10′W) and the southern Puget Sound area of Washington State, USA (47°13′N, 122°39′W) were live-captured using one of three methods. Swimming pups were captured using a seine net method [23] or an entanglement net method [24], while hauled-out pups were captured by hand [25].

Pups were manually restrained and blood samples were taken from the extradural vein using a Vacutainer blood collection system and heparinized plasma separation tubes (Becton Dickinson, Franklin Lakes, NJ, USA). The blubber biopsy site was cleansed with Hibitane disinfectant (Ayerst Laboratories, Montreal, QC, Canada), Betadine® (Purdue Frederick, Pickering, ON, Canada), and 95% isopropyl alcohol (Rhone Merieux, Victoriaville, QC, Canada) and 0.5 to 0.7 ml of Lidocaine (MTC Pharmaceuticals, Cambridge, ON, Canada) was administered intradermally. A 1-cm incision was made using a number 11 scalpel blade (Becton Dickinson) and a 150- to 200-mg blubber biopsy was taken using an Acu-Punch 6-mm biopsy sampler (Acuderm, Ft. Lauderdale, FL, USA) and wrapped in hexane-rinsed aluminum foil. The area was then cleansed again with Betadine and a topical anesthetic was administered (Xylocaine®, Astra Pharma, Mississauga, ON, Canada). Animals were weighed, sexed, measured for length and axillary girth, assessed for general body condition, and an interdigital tag was attached to the right hind flipper for identification purposes. Seals were then released at the site of capture within 30 min of the initial capture time, except fasted pups from British Columbia, which were used for a short-term captive study and later released. All blood and blubber samples were kept cool (4°C) and protected from light until arrival at the laboratory within 4 to 8 h of sampling. Blood tubes were centrifuged at 400 g for 20 min at 4°C, and plasma was aliquotted into 2-ml cryovials. Blubber and plasma samples were stored at −80°C until analysis.

**Analysis**

**Organochlorines.** Blubber samples were analyzed for congener-specific PCBs, PCDDs, and PCDFs by the Department of Fisheries and Oceans Regional Dioxin Laboratory at the Institute of Ocean Sciences in Sidney, British Columbia, Canada. Details on the sample extraction and clean-up procedures, instrumental analysis conditions used, quantification protocols, criteria used for congener identification, and quality assurance/quality control measures undertaken for the analysis of all analytes of interest are described elsewhere [26,27]. Toxic equivalency quotients (TEQs) were calculated using the most recent World Health Organization toxic equivalency factors [16].

**Total circulatory retinol.** Methods for the quantification of total circulatory retinol were adapted from a review article on vitamin A analysis [28]. Briefly, 200 µl of seal plasma was spiked with 0.1 µg of retinyl acetate (Sigma, St. Louis, MO, USA), which was used as the method internal standard. Using a liquid-liquid extraction method, samples were deproteinized with one part methanol, and retinoids were extracted twice using two partshexane. The methanol/hexane/sample mixture was centrifuged at 2,000 g, and the pooled organic layers were evaporated using a gentle stream of nitrogen. Retinoids were resuspended in 1 ml methanol and analyzed, in triplicate, using a reverse-phase high performance liquid chromatography system (Beckman System Gold Solvent Module 126, Fullerton, CA, USA) equipped with ultraviolet detection (Beckman System Gold Detector Module 166). A 20-µl sample volume was injected into the system and separated using a Vydac C18 column (0.46 × 25 cm, 5 µ) coupled to a guard column (0.46 × 1.0 cm, 5 µ) (Vydac, Hesperia, CA, USA). Retinol was eluted isocratically with a 90% methanol mobile phase, at a flow rate of 1 ml/min, and detected at a measurement wavelength of 325 nm. Quantification of retinol was obtained from linear calibration curves established using authentic retinol standards (Sigma).

**Circulatory retinol bound to its transport proteins.** The concentration of retinol bound to its transport proteins as RBP-retinol and TTR-RBP-retinol were analyzed using a size-exclusion chromatography method established by Burri and Kutnik [29]. Briefly, seal plasma was diluted 10 times with saline and filtered using a 0.2-µm low protein binding syringe tip filter. The sample was then analyzed, in duplicate, using the previously described chromatography system equipped with a Hewlett Packard 1046A fluorescence detector (Avondale, PA, USA) and the previously described ultraviolet detector in line. A 100-µl aliquot of the sample was injected into the system and separated using a Beckman Ultraspherogel Size Exclusion Column 2000 (7.5 mm × 30 cm, 5 µ) coupled with a guard column (7.5 mm × 4 cm, 5 µ) (Phenomenex, Torrance, CA, USA). The TTR-RBP-retinol and RBP-retinol were eluted isocratically with a pH 6.5 mobile phase (1 mM ethylenediaminetetraacetic acid, 2 mM β-mercaptoethanol, and 150 mM sodium phosphate) at a flow rate of 0.5 ml/min [29]. The TTR-RBP-retinol and RBP-retinol complexes were fluorescence measured using an excitation wavelength of 280 nm and emission wavelength of 330 nm before being detected at a measurement wavelength of 280 nm. Quantification of TTR-RBP-retinol and RBP-retinol complexes was obtained from linear calibration curves established from authentic RBP standards (Sigma) incubated at 4°C for 18 h with 3× molar excess of authentic retinol and TTR standards (Sigma).

**Statistics**

Harbor seal pups were categorized by study group, reflecting geographical origin and nursing status. The three study groups were (1) Washington State animals, which were recently weaned pups that were captured, sampled, and released in 1996 as part of an ongoing ecotoxicological study with the Washington Department of Fish and Wildlife (n = 16), and British Columbia animals, which were either (2) three- to four-week-old nursing pups that were captured, sampled, and re-
leased in 1997 as part of an ecotoxicological study \((n = 21)\) or \((3)\) three- to four-week-old nursing pups that were captured and fasted for 24 to 96 h before being sampled as part of a four-week-long immunotoxicological study in 1996 \((n = 24)\). Of the 61 harbor seal samples that were analyzed for retinol, PCBs, PCDDs, and PCDFs, four were statistical outliers and were not included in analysis (British Columbia nursing \(n = 20\), Washington State \(n = 15\), British Columbia fasted \(n = 22\)).

Among-group differences were assessed using a one-way analysis of variance and, when significant, a Tukey’s honest significant difference test was used post hoc to determine which study group differed. Regression analysis was used when data was analyzed as independent, ungrouped sets of data, and multiple regression analysis was used if more than one correlation was found to be significant. All data were analyzed using the statistical package Statistica© (StatSoft, Tulsa, OK, USA).

**RESULTS**

The concentration of total circulatory retinol ranged from 68 to 619 \(\mu g/L\). Harbor seal pups from Washington State had significantly lower levels of total circulatory retinol than pups from British Columbia (nursing and fasted) and fasted pups from British Columbia had significantly lower levels than nursing pups from British Columbia (Table 1). In all harbor seal pups analyzed, a positive correlation was measured between total circulatory retinol and body weight (i.e., age) in the dominant class of chemical (Table 2), and PCB-153, -138, -180, -99, -187, and -110 were found in the highest concentrations. The \(\Sigma PCBs\) were significantly correlated with the \(\Sigma TEQs\) for all harbor seal samples \((p < 0.01, r^2 = 0.950)\). Contaminant levels \((\Sigma PCB\) and \(\Sigma TEQ)\) did not correlate with body weight (i.e., age) in any of the study groups.

Contaminant levels \((\Sigma TEQ)\) were positively correlated with both total and unbound (free) circulatory retinol levels in recently weaned Washington State pups and in fasted pups from British Columbia (Figs. 1 and 2). However, there were no detectable effects of contaminants on total or free circulatory retinol levels in nursing pups from British Columbia.

In order to exclude the possible confounding factor of age, we analyzed each group (nursing, fasted, and weaned) separately to determine whether body weight (i.e., age) correlated with total circulatory retinol. We did not detect an effect of body weight on total circulatory retinol levels in recently weaned Washington State harbor seal pups or nursing pups from British Columbia. Fasted pups from British Columbia did show a significant positive correlation between total circulatory retinol and body weight \((p < 0.01, r^2 = 0.301; \text{data not shown})\). However, multivariate regression analysis indicated that this relationship was not significant when looking in all three groups, irrespective of contaminant levels and nursing status (Table 1). Over 95% of bound circulatory retinol existed as TTR-RBP-retinol.

Harbor seal pups from Washington State were more contaminated than British Columbia pups (nursing and fasting), as \(\Sigma PCBs\) were six times greater and the sum of 2,3,7,8-tetrachlorodibenzo-p-dioxin toxic equivalency quotients \((\Sigma TEQs)\) were three times greater in Washington State pups (Table 2). Of the contaminants measured, PCBs represented the dominant class of chemical (Table 2), and PCB-153, -138, -180, -99, -187, and -110 were found in the highest concentrations. The \(\Sigma PCBs\) were significantly correlated with the \(\Sigma TEQs\) for all harbor seal samples \((p < 0.01, r^2 = 0.950)\). Contaminant levels \((\Sigma PCB\) and \(\Sigma TEQ)\) did not correlate with body weight (i.e., age) in any of the study groups.

**Table 1. Circulatory vitamin A (retinol) levels in harbor seal pups from Washington State, USA, and British Columbia, Canada**

<table>
<thead>
<tr>
<th></th>
<th>Washington State (recently weaned)</th>
<th>British Columbia (fasted)</th>
<th>British Columbia (nursing)</th>
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<tr>
<td></td>
<td>(n = 15)</td>
<td>(n = 22)</td>
<td>(n = 20)</td>
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<tr>
<td>Total plasma retinol ((\mu g/L))</td>
<td>231.5 ± 39.9 A,B (\text{(68–593)})</td>
<td>343.5 ± 14.7 C (\text{(210–461)})</td>
<td>482.1 ± 26.1 (\text{(303–619)})</td>
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<tr>
<td>Retinol bound to transport</td>
<td>97.1 ± 3.0 (\text{(82–119)})</td>
<td>100.8 ± 2.9 (\text{(62–121)})</td>
<td>105.5 ± 3.0 (\text{(78–136)})</td>
</tr>
<tr>
<td>Proteins (TTR and RBP) ((\mu g/L))</td>
<td>52.7 ± 3.4 (\text{(28.0–83.9)})</td>
<td>48.2 ± 3.0 (\text{(20.3–4.7)})</td>
<td>44.1 ± 4.1 (\text{(15.7–82.2)})</td>
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*Values are mean ± standard error of mean (SEM) with range in parentheses; A = significantly different from fasted pups (Tukey’s post-analysis of variance [ANOVA] test; \(p < 0.05\)); B = significantly different from nursing pups (Tukey’s post-ANOVA test; \(p < 0.01\)); C = significantly different from nursing pups (Tukey’s post-ANOVA test; \(p < 0.05\)).

**Table 2. PCB, PCDD and PCDF levels in blubber biopsies of harbor seal pups from Washington State, USA, and British Columbia, Canada**

<table>
<thead>
<tr>
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<th>British Columbia (fasted)</th>
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<tr>
<td></td>
<td>(n = 15)</td>
<td>(n = 22)</td>
<td>(n = 20)</td>
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<tr>
<td>(\Sigma PCBs) (ng/kg lipid)</td>
<td>15.4 ± 2.2 A,B (\text{(6.9–33.8)})</td>
<td>2.3 ± 0.2 (\text{(1.1–4.6)})</td>
<td>2.5 ± 0.3 (\text{(0.5–4.7)})</td>
</tr>
<tr>
<td>(\Sigma PCDDs) (ng/kg lipid)</td>
<td>149.6 ± 21.1 A (\text{(62.0–378.4)})</td>
<td>343.9 ± 24.7 B (\text{(190.8–677.8)})</td>
<td>187.4 ± 54.0 (\text{(44.5–1,151.9)})</td>
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<tr>
<td>(\Sigma PCDFs) (ng/kg lipid)</td>
<td>12.2 ± 1.6 (\text{(3.9–28.2)})</td>
<td>42.9 ± 25.3 (\text{(8.3–538.3)})</td>
<td>20.3 ± 4.2 (\text{(0.0–75.2)})</td>
</tr>
<tr>
<td>(\Sigma TEQ) (ng/kg lipid)</td>
<td>154.0 ± 15.8 A,B (\text{(80.7–281.8)})</td>
<td>52.7 ± 3.4 (\text{(28.0–83.9)})</td>
<td>44.1 ± 4.1 (\text{(15.7–82.2)})</td>
</tr>
</tbody>
</table>

*Values are mean ± standard error of mean (SEM) with range in parentheses; A = significantly different from fasted pups (Tukey’s post-analysis of variance [ANOVA] test; \(p < 0.01\)); B = significantly different from nursing pups (Tukey’s post-ANOVA test; \(p < 0.01\)).

*PCB = polychlorinated biphenyl; PCDD = polychlorinated dibenzo-p-dioxin; PCDF = polychlorinated dibenzofuran.
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Fig. 1. Total circulatory retinol levels in harbor seal pups as a function of total toxic equivalents (TEQ) measured in their blubber. Recently weaned pups from Washington State, USA (open symbols; $p < 0.01$, $r^2 = 0.66$), and fasted pups from British Columbia, Canada (filled symbols, $p < 0.05$, $r^2 = 0.24$), showed significant positive correlations. Due to differences in nursing status, vitamin A levels varied between the two groups.

Fig. 2. Concentration of retinol circulating free in the blood of harbor seal pups (unbound by transport proteins) as a function of total toxic equivalents (TEQ) measured in their blubber. Recently weaned pups from Washington State, USA (open symbols; $p < 0.01$, $r^2 = 0.64$), and fasted pups from British Columbia, Canada (filled symbols; $p < 0.05$, $r^2 = 0.26$), showed significant positive correlations. Due to differences in nursing status, vitamin A levels varied between the two groups.

at all three factors (total circulatory retinol levels, body weight, and $\Sigma$TEQ).

**DISCUSSION**

During lactation, female harbor seals mobilize a large proportion of their blubber reserves to produce an energy-rich milk comprised of almost 50% fat [30]. Although this milk contains energy and nutrients that are essential for pup survival, it also serves as a medium for the transfer of lipophilic organochlorines, including PCBs, PCDDs, and PCDFs [31,32]. As a result, harbor seal pups can be exposed to relatively high concentrations of contaminants during their estimated four- to six-week nursing period. This exposure comes at a time when animals are thought to be relatively sensitive to the toxic effects of contaminants and when maternally derived vitamin A is essential for growth and development [10].

While single-chemical experiments can determine the toxic effects of organochlorines in laboratory animals, complex environmental mixtures of contaminants make wildlife studies much more difficult to interpret. The $\Sigma$TEQ, which is significantly correlated with $\Sigma$PCB, represents a measurement of contaminant exposure that simplifies environmental mixtures on the basis of total dioxin-like toxicity [16]. In the present study, harbor seal pups from Washington State were significantly more contaminated ($\Sigma$TEQ and $\Sigma$PCB) than harbor seal pups from British Columbia, presumably as a result of the high industrial activity in the Puget Sound area of Washington State. In addition to being more contaminated, Washington State harbor seal pups had significantly lower circulatory vitamin A levels than the relatively uncontaminated pups from British Columbia (nursing and fasted). This observation appears consistent with the results of other studies, where vitamin A levels in contaminated groups of seals have been lower than levels in relatively uncontaminated groups of seals [8,9]. However, because the lower circulatory retinol levels observed in our Washington State seals may have been attributed to differences in nursing status [25], we further analyzed our data as three ungrouped sets of data points. This revealed that total circulatory retinol levels within (nonnursing) study groups were positively correlated with contaminant exposure ($\Sigma$TEQ). Similar relationships were observed in both the contaminated, recently weaned pups from Washington State as well as the less contaminated, fasted pups from British Columbia, suggesting that even low levels of contaminants are affecting retinoid dynamics in free-ranging harbor seals. The lower overall vitamin A levels observed in the Washington State study group likely reflected their weaned status. Although negative correlations between contaminant and retinoid concentrations have been observed in other seal studies [20,33], the effects of nursing status and nutritional state may have interfered with data interpretation, as both are known to affect total circulatory retinol levels in seals [25].

Although all harbor seal pups from British Columbia were of similar ages, contaminant levels, and geographical origin, we did not detect an effect of contaminants on total circulatory retinol levels in nursing harbor seal pups from British Columbia. This suggests that nursing may represent a confounding factor in determining the effects of contaminant exposure on vitamin A dynamics. This may be due to the fresh input of large quantities of maternally derived vitamin A in the milk. This potentially confounding effect of maternally derived vitamin A was eliminated in our weaned (Washington State) and fasted pups (British Columbia), thereby allowing a better assessment of contaminant-related effects in the pup.

Harbor seal pups that exhibited a positive correlation between contaminant exposure and total circulatory retinol also had elevated levels of retinol circulating free in the blood, unbound to TTR and/or RBP. This relationship may have resulted from a disruption of the vitamin A transport complex, as laboratory studies have shown that some PCB hydroxy metabolites can bind TTR, disrupting the circulatory transport of retinol [19]. Although PCB hydroxy metabolites have previously been identified in blood samples taken from adult seals [34], only limited amounts are thought to be lactationally transferred to offspring in rodents [35,36], and the immature seal pup liver has a limited enzymatic ability to generate its own metabolites [37]. This suggests that pups, especially nonnursing pups, might have relatively small amounts of hydroxy metabolites circulating in the blood. If this small amount were
to disrupt the vitamin A transport complex, we might expect less retinol to be in the form of TTR-RBP-retinol and lower concentrations of RBP-retinol, RBP, and retinol in the blood because they are more readily filtered by the kidney and excreted [19]. In the present study, however, concentrations of retinol bound and regulated by its transport proteins (as TTR-RBP-retinol and RBP-retinol) remained constant at all levels of exposure and total circulatory retinol levels were positively correlated with contaminant exposure. This suggests that the principal mechanism of contaminant-induced vitamin A toxicity in our study was likely not mediated by a metabolite-related disruption of the vitamin A transport complex.

In laboratory studies, organochlorine exposure has been shown to inhibit the intestinal absorption and hepatic storage of newly ingested vitamin A [2,4] and can also lead to a mobilization of existing vitamin A stores in the liver and extrahepatic tissues [4,6,7]. These effects, which are thought to result from contaminant-induced alterations in the activity of enzymes involved in vitamin A metabolism [17], can result in decreased liver vitamin A stores and increased circulatory retinol concentrations [2,5,6]. Although liver vitamin A levels were not measured in this study, it is possible that the contaminant-associated increases observed in circulatory retinol levels in our study seals were due to a mobilization of liver vitamin A stores into the circulation.

Laboratory-based studies suggest that a contaminant-induced reduction of liver vitamin A stores can occur rapidly [2,7] and at relatively low contaminant levels [38] in animals exposed to PCBs and PCDDs orally [3–6,39] or via the milk [36,40]. Wildlife exposed to complex environmental mixtures of contaminants have also exhibited severe reductions in liver vitamin A stores, with increased liver retinol:retinyl ester ratios and increased circulatory retinol:retinyl ester ester ratios suggesting a mobilization of liver stores into the circulation [21,22]. However, when depletion of liver vitamin A stores becomes acute, circulatory retinol levels will eventually drop [15]. This is consistent with observations of contaminant-related reductions in circulatory vitamin A levels in studies of stranded and malnourished pups [33], which likely have limited liver reserves of vitamin A, and of adult seals chronically exposed to contaminants via the diet [8,9]. In our study of healthy young harbor seal pups, the positive correlations observed between contaminants and vitamin A may reflect an acute exposure to fat-soluble contaminants in the mothers’ milk, which may ultimately result in a depletion of liver vitamin A stores and reduced circulatory retinol levels.

In summary, we found that circulatory vitamin A levels were positively correlated with contaminant exposure in free-ranging harbor seal pups that were not nursing. As a result, vitamin A may represent a sensitive, noninvasive indicator of contaminant exposure, as vitamin A dynamics were affected even at relatively low levels of exposure. However, age and nursing status represent important considerations in our study design, as these factors affected circulatory vitamin A levels irrespective of an effect of contaminant exposure. Although it is possible that the circulatory transport of vitamin A was disrupted by organochlorine hydroxy metabolites, we found no evidence to support this in our study of young harbor seal pups. The increased circulatory retinol levels observed in our study seals suggest a contaminant-induced mobilization of liver vitamin A stores, but an examination of the relationship between circulatory, hepatic, and tissue vitamin A levels in exposed seal pups would yield more conclusive insight into this observation. This contaminant-related loss of vitamin A during a period of rapid growth and development could adversely affect the health of young harbor seals even in relatively unpolluted coastal regions.

Acknowledgement—We would like to thank Brian Beck, Jason Ngui, Paul Cottrell, Peter Olesiuk, Dyanna Lambourne, and Tom Smith for their valuable assistance in the field. We also thank Betty Jane Burri and Terry Neidlinger for their advice in setting up the binding protein assay. Special thanks to Neil Dangerfield and Derek Smith for their analytical support throughout the project. This work was supported by the Fisheries and Oceans Canada Toxic Chemicals Program, Ottawa, Ontario, Canada.

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