Exposure of Pacific Herring (*Clupea pallasi*) to Persistent Organic Pollutants in Puget Sound and the Georgia Basin

Sandra M. O'Neill *and* ***James E. West***
Washington State Department of Fish and Wildlife

Abstract

The Puget Sound Ambient Monitoring Program has monitored spatial and temporal trends of toxic contaminants in Puget Sound fishes from 1989 to the present. A pilot study on the feasibility of adopting Pacific herring (*Clupea harengus*) as an additional indicator species was conducted in 1995, and regular monitoring of herring begun in 1999. Pilot sampling of toxic contaminants in herring eggs was also begun. We have focused on two classes of toxic contaminants that accumulate in fish tissues, (polychlorinated biphenyls (PCBs), and pesticides (DDT, its metabolites, and hexachlorobenzene (HCB)), and one class that does not accumulate in fish tissues (polycyclic aromatic hydrocarbons (PAHs)). PCBs and pesticides were measured as concentrations from composites of whole fish. Recent exposure to PAHs was estimated from composites of fish bile, by measuring concentration of PAH-metabolites known as Fluorescing Aromatic Compounds (FACs). We also measured PAH concentrations in eggs-recently- laid by two distinct spawning stocks in Puget Sound, one near an oil refinery and one away from suspected oil sources.

Total PCBs in whole herring bodies in two Central/South Puget Sound locations ranged from 125 to 350 µg/kg, and were significantly greater than three northern locations (all less than 125 µg/kg). ppDDE comprised the bulk of the body burden of DDT and DDT-metabolite compounds in herring, with the greatest concentrations observed from the central Sound station (40 to 175 µg/kg. Herring from all other locations had concentrations less than 40 µg/kg. HCB was detected often, but in low concentrations (<1.8 µg/kg).

Biliary FACs (metabolites of benzo-a-pyrene, naphthalene, and phenanthrene) were significantly greater in herring from a central Sound location than northern and a southern Sound locations. We observed no correlation between FAC concentration and biliary protein.

PAH and PAH-homologue compounds were not detected from herring eggs laid by the Port Gamble spawning stock at two locations (away from an oil refinery), however those eggs were only one-day-old. Eggs laid by the Cherry Point spawning stock at the three sites (near an oil refinery) were 7 to 8 days old, which confounded valid comparison with the Port Gamble egg samples. Eggs from two of the three Cherry Point sites had detected PAHs (dibenzothiophene, acenaphthene, naphthalene, 2-methylnaphthalene, and C-1 naphthalenes) ranging in concentration from 1.0 to 3.5 µg/kg (wet wt.) Presence of some of these compounds is consistent with exposure to Alaska north slope crude oil, (which is refined nearby) however, concentrations were below a published lowest observable effects concentration (LOEC).

Introduction

Pacific herring (*Clupea pallasi*) are important prey to many other fish species, seabirds and marine mammals (Hart 1973) Consequently, the health of these higher trophic levels is linked to the health of herring in the Puget Sound and Georgia Basin ecosystem. Adult Pacific herring are pelagic and wide ranging but they move into the shallow, nearshore habitat to spawn on marine vegetation or other structures. Fertilized eggs hatch in about 14 days and the pelagic larvae gradually move from the nearshore habitat to the open water column (Hart 1973). Within Puget Sound, these spawning aggregations are managed as discrete stocks by the Washington Department of Fish and Wildlife (Lemberg and others 1997). In the late winter and early spring, large spawning aggregations of adult herring provide rich sources of prey to various piscivorous fishes and marine birds and mammals. Their freshly spawned eggs are also consumed by benthic fishes and seabirds. Larval and juvenile herring are also key components of the region's marine food web (Hart 1973; Lemberg and others 1997).

Because of their importance to the food web, there is concern that if herring are exposed to toxic contaminants and accumulate them, much of the local food web could be affected. We conducted a pilot study on individual herring from Fidalgo Bay in 1995, demonstrating that adult herring accumulated polychlorinated biphenyls (PCBs), and low levels of mercury, arsenic and copper. In 1999 we initiated an on-going program to monitor contaminant levels in adult herring stocks from Puget Sound and the Georgia Basin. The objective was to measure contaminant exposure in whole bodies of adult herring from different spawning stocks to assess spatial variation in contaminant body burdens. We estimated exposure to bio-accumulative organochlorines by measuring whole body concentrations of PCBs, chlorinated pesticides (DDT and its metabolites) and hexachlorobenzene (HCB). Recent exposure to polycyclic aromatic hydrocarbons (PAHs), organic compounds that do not accumulate in fish, was monitored by estimating biliary concentrations of PAH-metabolites, measured as Fluorescing Aromatic Compounds (FACs). These organic pollutants were measured because many are lipophilic and would probably accumulate in fatty fish like herring.

Average contaminant exposure in adult spawning stocks should reflect environmental contamination from the geographic areas in which they reside. To minimize variation within and among sampling locations related to reproductive condition and fish age, chemical analysis was limited to composite samples of adult males, ranging in age from 2 to 4 years. Furthermore, because herring are a short-lived species and younger fish were selected, their contaminant loads reflect recent exposure to contaminants.

Materials and Methods

A rope trawl was used to collect adult herring from predictable pre-spawning aggregations. Sampling areas were selected to represent contaminant exposures in herring from a broad geographic range. In 1999 we sampled five spawning stocks: Denby/Hornby (Georgia Basin), Semiahmoo (northern Puget Sound), Cherry Point (northern Puget Sound), Port Orchard (central Puget Sound), and Squaxin Pass (southern Puget Sound). In 2000 we repeated our sampling on the Semiahmoo, Port Orchard and Squaxin Pass spawning stocks. Cherry Point fish spawn in April but all the other stocks complete their spawning by the end of February.

At each sampling location, we collected two groups of fish for chemical analyses: one for analysis of PCBs, DDTs, and HCB in whole fish and the other for analysis of PAH metabolites in bile. Up to 400 unsexed fish were selected in the field for whole body contaminant analysis and then taken to our lab, sexed, and scales were removed for age determination. Fifty males, aged 2 to 4 years, were selected and combined into 10 composite samples of 5 fish each. Fish to be used for bile FAC analyses were sexed in the field and bile was only extracted from adult males. Adequate bile samples were only collected in 2000. Ten bile composites samples were collected at each sampling location by combining equal amounts of bile from 5 to 9 individual fish.

PCBs, DDT/metabolites, and hexachlorobenzene were analyzed by high performance liquid chromatography (HPLC) with photo-diode array (Krahn and others 1994), and bile FACs were analyzed by HPLC with ultraviolet fluorescence (Krahn and others 1987).

Results

Organochlorines in whole bodies

Total PCBs in whole bodies of herring (Table 1) from Port Orchard and Squaxin Pass, representing the central and southern Puget Sound stocks were significantly higher than those from the three northern locations, Semiahmoo, Cherry Point, and Denby/Hornby (ANOVA , $p < 0.001$,; Tukey's post-hoc pairwise comparisons). Lipid content of the samples also varied significantly among spawning stocks (ANOVA, $p < 0.001$) confounding the interpretation of these results. However, for the three stocks whose lipid values were similar to each other, (Denby/Hornby, Semiahmoo, and Port Orchard; Tukeys), PCB concentrations were highest in herring from the central Puget Sound (Figure 1). The elevated PCB concentration for Squaxin Pass fish, the most southern Puget Sound stock, was probably in part related to the significantly higher lipid levels observed for that spawning stock. However, the few Squaxin Pass samples with lipid

levels that were similar to those from other locations had PCB concentrations that were more like those from Port Orchard than the northern Puget Sound and the Georgia Basin (Figure 1). Significantly lower lipid levels were observed in the Cherry Point herring than all other stocks (Tukey's post-hoc pairwise comparison) however, for those individual samples where lipid levels were similar to those from Georgia Basin and northern Puget Sound, PCB concentrations were also similar. We concluded that when differences in lipid content are accounted for, PCB concentrations were significantly higher in fish from the central and southern than the more northern locations.

Table 1. Mean (and standard deviations) organochlorine concentration (µg/kg wet weight) in Pacific herring stocks from the Georgia Basin and Puget Sound.

Stock	Mean Fish Age (years)	N	% Lipids Mean	Total PCBs Mean	Total PCBs s.d.	Total DDTs Mean	Total DDTs s.d.	HCB Mean	HCB s.d.
Denby/Hornby	2.8	10	5.19	17.0	4.0	18.0	7.3	1.33	0.47
Cherry Point	2.8	10	3.59	54.9	13.0	12.7	3.0	0.35	0.19
Semiahmoo	2.8	20	5.62	52.3	24.7	17.8	7.0	1.54	0.27
Port Orchard	2.8	20	6.52	200.2	48.7	47.2	39.4	1.43	0.18
Squaxin Pass	2.9	19	9.15	165.8	48.8	20.9	5.0	1.43	0.27

The highest concentration of total DDTs (represented mostly as ppDDE) was also observed in Port Orchard herring (Table 1). The significance of these results is unclear as the concentration of ppDDE in these fish was elevated in 1999 but less so in the 2000 samples. HCB was detected often, but in low concentrations. Significantly lower HCB concentrations were observed for the Cherry Point stock than for all other stocks (ANOVA, $p<0.001$; Tukey's post-hoc pairwwise comparison).

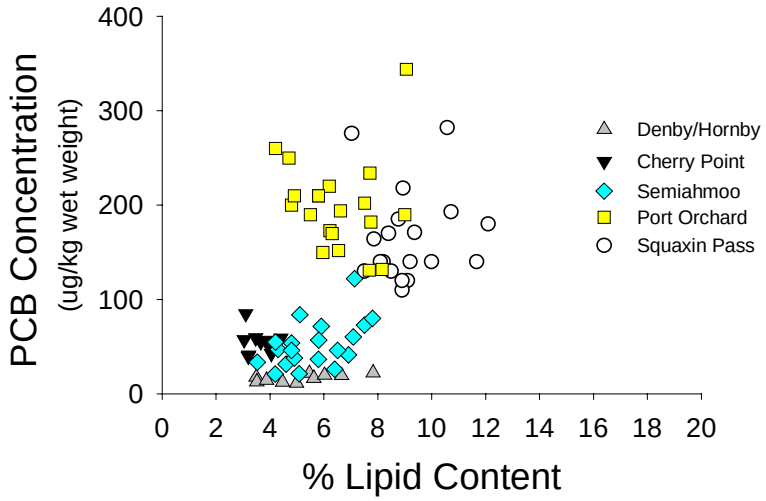

Figure 1. Relationship between lipid content and PCB concentration in whole bodies of Pacific herring from Georgia Basin and Puget Sound stocks.

PAH metabolites in bile

Biliary FACs (PAH metabolites expressed as equivalents of benzo-a-pyrene, and phenanthrene) were also highest in herring from Port Orchard in 2000 (ANOVA, $p<0.0001$; Tukey's post-hoc pairwise comparison). The Port Orchard bile samples also had significantly greater protein concentration (a measure of bile diluteness that usually correlates positively with FAC concentrations) possibly explaining the elevated FAC

levels. However, stock location was likely an important factor as well because for those individual samples where protein values were similar among stocks, Port Orchard FACs levels were always higher than those observed in Semiahmoo and Squaxin Pass fish (Figure 2).

Table 2. Mean (and standard deviation) PAH metabolite concentration in Pacific herring stocks from the Georgia Basin and Puget Sound.

		Protein (mg/ml)		FAC BaP (ng/g bile)		FAC PHN (ng/g bile)	
Stock	**N**	**Mean**	**s.d.**	**Mean**	**s.d.**	**Mean**	**s.d.**
Semiahmoo	10	6.62	2.63	224.6	68.5	22662	4074
Port Orchard	9	10.73	2.74	551.4	87.7	52237	4941
Squaxin Pass	10	5.77	2.86	305.0	91.4	26829	4955

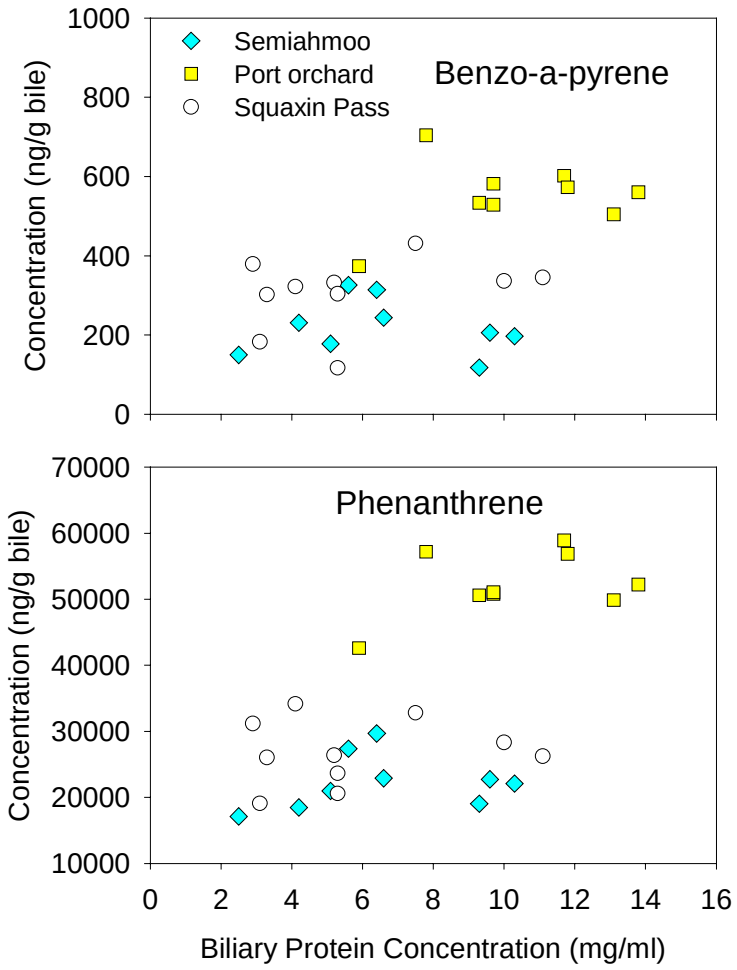

Figure 2. Relationship between concentration of biliary proteins and biliary FACs (estimated as equivalents of benzo-a-pyrene and phenanthrene) in Pacific herring stocks from the Georgia Basin and Puget Sound.

Discussion

In this study we documented that Pacific herring from the central and southern Puget Sound basins had higher body burdens of PCBs than fish from northern Puget Sound and the Strait of Georgia. We also observed higher PAH values in the herring from the central Puget Sound relative to all other locations. We observed higher DDTs concentration in fish from the Port Orchard stock; however, this was mostly due to higher values for the 1999 samples. Low HCB concentrations were observed for all stocks and were significantly lower for the Cherry Point stock.

The higher toxics levels in herring from the central basin are likely associated with its greater area of contaminated sediments. Polychlorinated biphenyls (PCBs) and poly-aromatic hydrocarbons (PAHs) are persistent organic pollutants that accumulate in marine sediments adjacent to urban and industrialized centers (Varanasi and others 1992). Most of the urban bays that have PCB and PAH contaminated sediments are located in the central Puget Sound basin (Everett Harbor, Elliott Bay, Sinclair Inlet, Eagle Harbor, and Commencement Bay)(Dutch and others 1993) (Long and others 2000). We suggest that PCBs are transferred from these contaminated sediments to the zooplankton in the central and southern basin by biological processes. For example, macro-invertebrates and fishes associated with contaminated sediments may accumulate PCBs that are then maternally transferred to their planktonic eggs and larvae, which in turn become part of the herring's pelagic food web.

These data suggest that PCBs and PAHs permeate through the food web of the largest basin of the Puget Sound ecosystem (the Whidbey/Central basin), and that PCBs may also be substantially present in the southern basin food web. Through our ongoing PSAMP studies, we have documented that wild coho salmon from southern Puget Sound have higher lipid-specific PCB concentrations than fish from northern Puget Sound, possibly the result of their longer residence in central and southern Puget Sound during their out- and in-migrations (O'Neill and others 1998). Similarly, the PSAMP Marine Mammal Component documented higher PCB concentrations in harbor seals in southern Puget Sound than in northern Georgia Strait (Calambokidis and others 1988) (Ross and others 1998). We suggest that prey of these animals, like Pacific herring, may be more contaminated in the more industrialized basins of central and southern Puget Sound than in the less developed basins of northern Puget Sound and Georgia Strait.

Based on a recent Adverse Effects Threshold developed by National Marine Fisheries Service (Meador 2000), most herring from Puget Sound are not likely adversely affected by exposure to PCBs. All of the PCB exposures were well below the 50^{th}-percentile threshold concentration (~12 ug/g lipid) that is associated with adverse effects. Based on the 10^{th}-percentile concentration that is associated with adverse effect, we would conclude that herring from the central Puget Sound are exposed to PCB concentrations that are potentially damaging.

Future analyses should allow us to more accurately define spatial patterns in toxics exposure in Pacific herring stock in Puget Sound and the Georgia Basin. Ongoing monitoring should help to define the lipd:PCB, lipid:DDT, lipid:HCB and protein:bile FAC relationships, allowing better comparisons among sampling locations. We have also initiated a study to assess whether organic contaminants are accumulated in Pacific herring eggs.

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