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Monitoring xenoestrogen exposure and endocrine disruption in a Puget Sound benthic flatfish, English sole (*Parophrys vetulus*)

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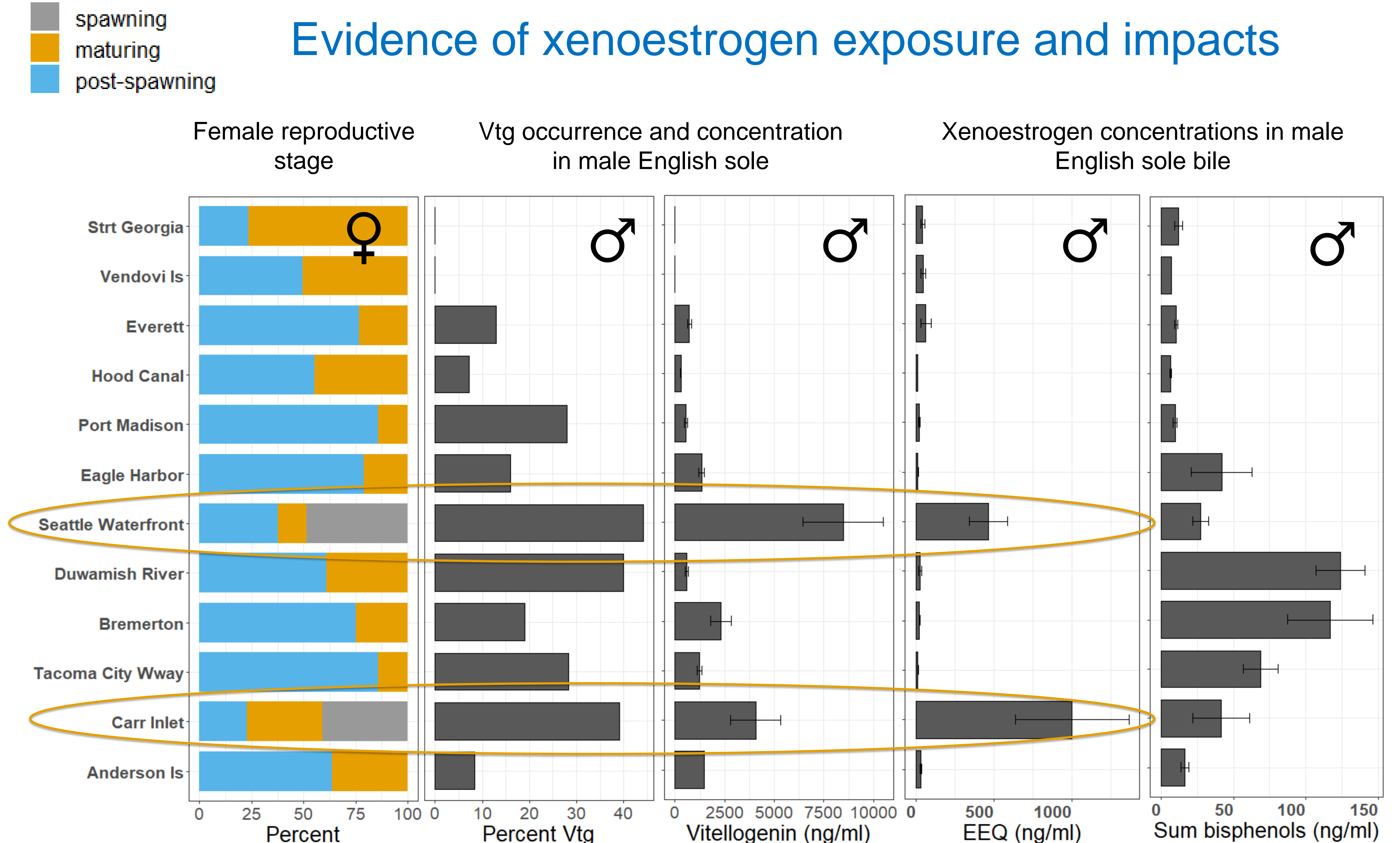
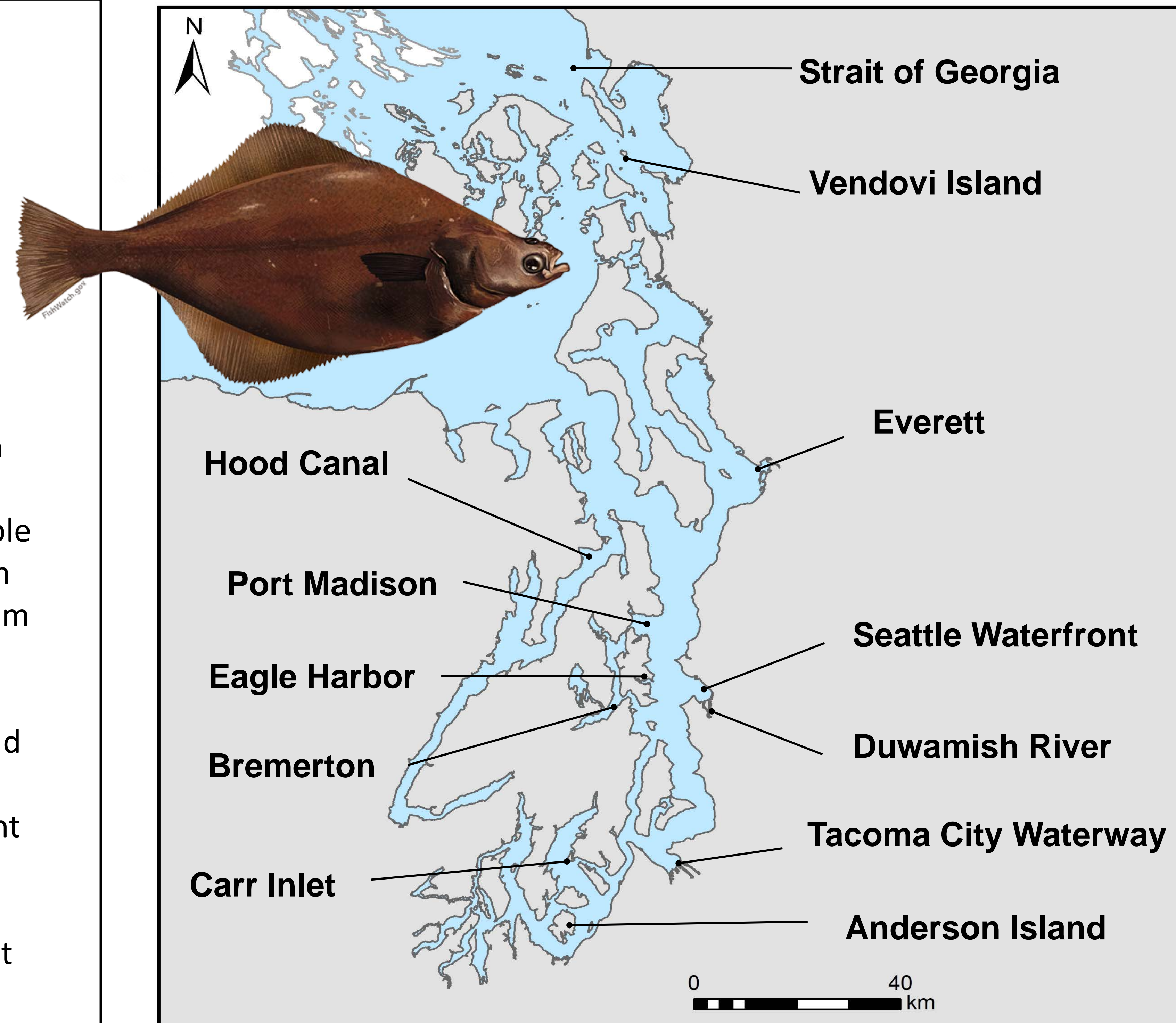
Harding, Dr. Louisa, "Monitoring xenoestrogen exposure and endocrine disruption in a Puget Sound benthic flatfish, English sole (*Parophrys vetulus*)" (2022). *Salish Sea Ecosystem Conference*. 116. <https://cedar.wwu.edu/ssec/2022ssec/allsessions/116>

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Monitoring xenoestrogen exposure and endocrine disruption in a Puget Sound benthic flatfish, English sole (*Parophrys vetulus*)

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Introduction: Xenoestrogens, including natural estrogens and synthetic chemicals that mimic estrogens such as bisphenol-A, can disrupt endocrine functions and reproduction in exposed fish. Vitellogenin (Vtg) is an egg-yolk protein precursor synthesized in the liver of female fish in response to naturally produced estrogens during their normal reproductive cycle. However, Vtg can be abnormally induced in male fish if they are exposed to xenoestrogens, making this protein a suitable and widely used biomarker of xenoestrogen exposure. In the late 1990s, researchers from WDFW and NOAA observed evidence of xenoestrogen exposure in English sole, including vitellogenin induction in males and altered reproductive timing (Johnson et al., 2008). This research investigated the current geographic extent and severity of xenoestrogen exposure in English sole, and which chemicals may be having the greatest impact.



Methods: Adult English sole were sampled from 12 monitoring locations throughout Puget Sound in May of 2017 and 2019. Vitellogenin levels were measured in male fish plasma using a commercial ELISA kit (TECO, TE1042). Gonads were collected for histological analysis to confirm sex and determine reproductive stage. Lastly, bile was collected and analyzed for xenoestrogens including 3 natural steroid estrogens and 5 bisphenols as described in da Silva et al., 2013. An estradiol equivalency (EEQ) was calculated as the sum of steroid estrogens measured in male English sole bile multiplied by their estradiol equivalency factor as follows: $EEQ = \text{estradiol} * 1 + \text{estrone} * 0.11 + \text{estriol} * 0.11$.

Acknowledgements: Thanks to Kurt Dobszinsky and the crew of the Chasina for assistance with fish collection, Denis da Silva for bile xenoestrogen analysis, Mark Myers for histological analysis, and to Lyndal Johnson and other past WDFW and NOAA staff who contributed to this work.

Literature cited: da Silva et al., 2013. Chemosphere 92, 1550-1556. Johnson et al., 2008. Aq. Tox. 88, 29-38.

For more information, visit: <https://wdfw.wa.gov/species-habitats/science/marine-toxics>;
<https://vitalsigns.pugetsoundinfo.wa.gov/VitalSign/Detail/11>

Results and Conclusions:

- Vtg expression in male English sole was widespread throughout most of Puget Sound consistent with previous work by Johnson et al., (2010).
- Altered reproductive timing in females, high Vtg occurrence in males, and high plasma Vtg concentrations all co-occurred at Seattle Waterfront and Carr Inlet providing good evidence of xenoestrogen exposure and endocrine disruption at these two locations.
- Male fish collected from Seattle Waterfront and Carr Inlet also had high levels of steroid estrogens in their bile, suggesting steroid hormones from sewage are a more likely cause of endocrine disruption in English sole than bisphenols.
- These results suggest that endocrine disruption may occur in English sole anywhere sewage enters Puget Sound, through leaky septic systems (rural) or wastewater treatment plant effluent and combined sewer overflows (urban) – a problem that could increase as population growth continues throughout the region.