The use of image-analysis software in evaluating spawning strategy and fecundity rates of Walleye pollock, *Gadus chalcogrammus*

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Introduction:

Walleye pollock populations are managed by applying a harvest rate toward an assessment model estimate of spawning stock biomass (SSB). SSB is used to implement the allowable biological catch (ABC) control rule and it is assumed that SSB is proportional to total egg production (fecundity). Because the ABC control rule requires that fishing mortality be reduced linearly below an inflection point (B40% the biomass when fishing at F40%), yield can be highly dependent on spawning biomass. Spencer and Dorn used simulation modeling of the Gulf of Alaska walleye pollock stock assessment to show an example of how reproductive dynamics may affect stock productivity. They found that the effect of weight-specific relative fecundity (i.e., increasing fecundity per unit body mass with weight) was to increase the estimate of F_{MSY} (fishing mortality rate at maximum sustainable yield) relative to an estimate using SSB as reproductive potential.

Goal of this project: Estimate variability in fecundity and examine functional relationships between fecundity and stock biomass and environmental factors so that fecundity can be treated dynamically in stock assessment

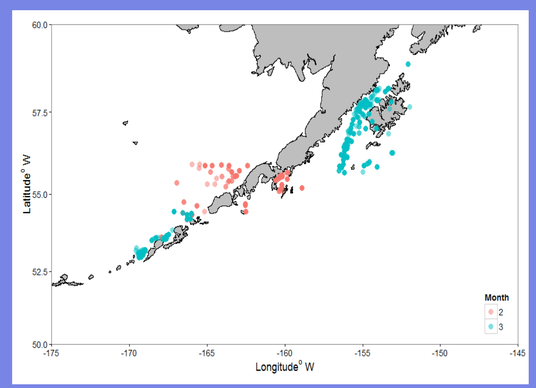

Figure 1 Specimen collection locations

Methods

Length and weight data were collected from ~1,350 ovaries from 1996-1998, 2000-2007 and 2012-2015. Pollock were caught during Alaska Fisheries Science Center pollock acoustic cruises in the Gulf of Alaska, Aleutian Islands, and Eastern Bering Sea (Figure 1) on the R/V Miller Freeman from 1996-2007, and on the R/V Oscar Dyson in 2012-2015. The majority of fish were caught from the beginning of February to the third week in March. Ovaries were assigned a gross anatomical maturity stage and preserved individually in cloth bags in 10% formalin. Specimen collections for fecundity analysis focused on ovaries in the Prespawning stage, just prior to spawning.

Histological evaluation

Tissue samples from each specimen were processed by a professional histology laboratory and evaluated microscopically. Specimens that show signs of prior spawning such as hydration or post-ovulatory follicles (POF) were omitted from this analysis (see histology images 1-4).

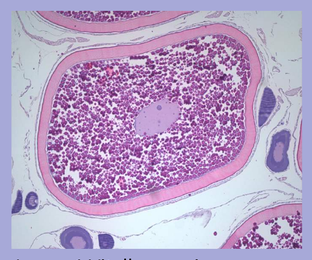

Image 1 Vitellogenesis

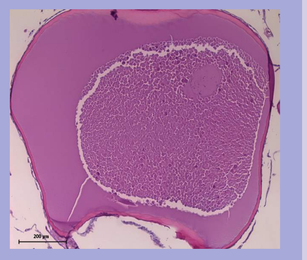

Image 2 Hydration

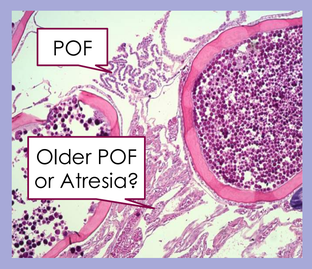

Image 3 POF

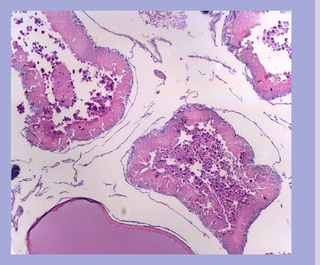

Image 4 Atresia

Fecundity

The gravimetric method was primarily used to evaluate fecundity. This method estimates fecundity by extrapolating a counted and weighted subsample out to the whole ovary weight. We also adapted the auto diametric method (Thorsen and Kjesbu, 2001) which establishes a relationship between the mean oocyte diameter and density (the number of oocytes per weighed subsample). This is done quickly through an image analysis program that digitizes the image of the oocyte sample.

Image J Freeware

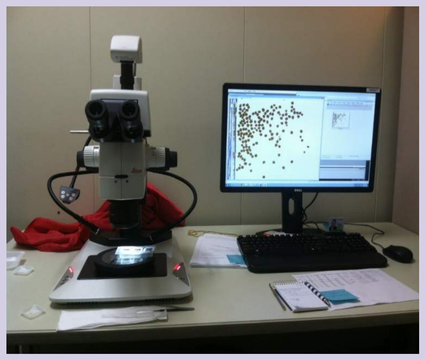

Image 5. Leica M125 stereomicroscope with DFC295 Color Camera and LED5000 SLI spotlight illuminator. Image 6 (to the right). shows oocyte water wells

Image J freeware was used to determine oocyte counts and the mean oocyte diameter of each specimen. A stereoscope with a digital camera, and illumination adjusted to give crisp edges was used to obtain accurate oocyte measures (Image 5). A subsample was weighed and then placed in a water filled well where oocytes were separated as much as possible (Image 6). Program digitized images were analyzed to count and measure individual oocytes (Image 7). Red lines represent mean diameter measures and numbers are the counts associated with each measured oocyte. The yellow circles highlight measurements of clumped oocytes or tunica identified by the program as a single shape. Such errors were easily deleted within the program. Measurements obtained include total oocyte count, average diameter and roundness, and standard deviation. These mean oocyte counts, along with a subsample weight, were used for gravimetric fecundity estimates

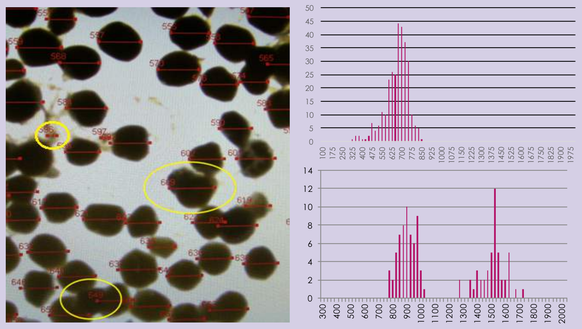

Image 7. Image J digitized image with counts and average mean. Yellow circles identify reading errors which are corrected manually. Figure 2. Histograms show oocyte development during maturation. Oocytes above 1000um were found to be hydrated.

Results

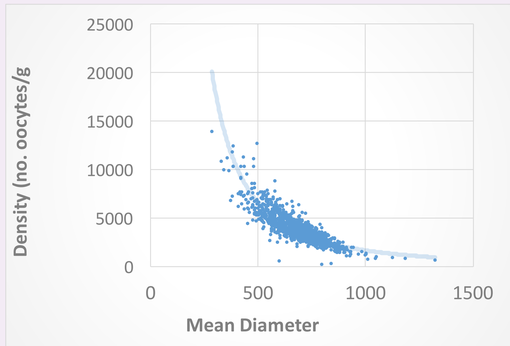

Figure 3. A regression curve with mean diameter from the data showing density decreasing as oocyte diameter increases. Once this relationship is established, fecundity can be found quickly through microscopic imaging and image analysis processing while avoiding chemical preservatives and long processing times.

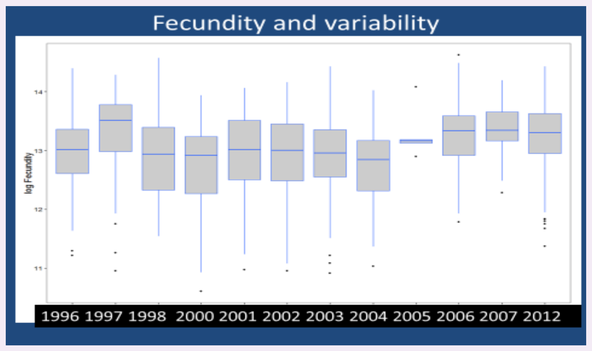

Figure 4. Fecundity estimates are fairly steady by year, with a few notable exceptions; 1997,2006, 2007 and 2012

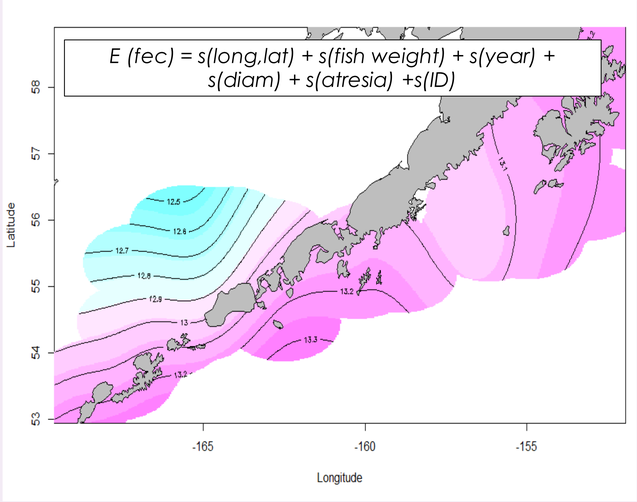

Figure 5. Partial residuals of a (GAM) are mapped to show the size distribution of fecundity by capture location. Warm colors = higher levels of fecundity; cool colors = lower levels of fecundity. Estimated fecundity (log transformed) is a product of the sample location, weight, the year of sampling, the mean egg diameter, an index of atresia (histological evaluation). The overall trend shows higher fecundity levels in Gulf of Alaska near Shelikof Strait and along the Aleutian Islands, the Eastern Bering Sea shows lower fecundity levels.

References

Thorsen, A. and Kjesbu, O.S. (2001). A rapid method for estimation of oocyte size and potential fecundity in Atlantic cod using a computer-aided particle analysis system. Journal of Sea Research 46, 295-308.

Spencer, P. D., and M. W. Dorn. 2013. Incorporation of weight-specific relative fecundity and maternal effects in larval survival into stock assessments. Fisheries Research 138:159–167.

Acknowledgments

In memory: Bern Megrey

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NOAA Fisheries and the Environment