Using Image Analysis Software to Assess American Eel (*Anguilla rostrata*) Elver Development

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Introduction

American eel (Anguilla rostrata) is a catadromous fish species that spawns in the Sargasso Sea. Larvae are transported to continental North America via the Gulf Stream and coastal currents. The run-timing (Figure 1) and run strength (Figure 2) of post-larval glass eels -or elvers- to non-tidal freshwater habitat varies among years and is influenced by a number of factors including time of arrival to the coastal zone, tidal influences, and local river conditions (e.g., flow, temperature).

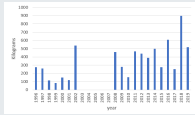


Figure 1. Total annual East River run-size (Kg) by year

Elvers have been fished commercially in Atlantic Canada since the 1980s principally to supply the Asian live eel market. Since 1996 Fisheries and Oceans Canada (DFO) and commercial license holders have collaborated on monitoring the run-timing, run-strength, and physiological traits of the annual runs of elvers to East River-Chester, Nova Scotia (Figure 6).

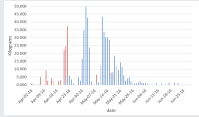


Figure 2. Total daily catch (Kg) versus Date in 2016

Both elver length and elver weight decline over the course of the annual runs. The decrease in body size affects the number of elvers contained within sold catches that are measured in terms of weight (Figure 3). Paradoxically, and while seemingly declining in body condition elver runs also exhibit increasing pigmentation and higher incidence of feeding, which are suggestive of increasing fitness. How these ecological and physiological factors influence recruitment are not currently fully understood. Understanding the physical development is important to managing the elver fishery.

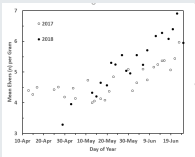


Figure 3. Change in elvers per gram with time for the 2017 and 2018 run years

Degree of pigmentation is generally regarded as a measure of physiological development for elvers. Pigmentation is currently assessed using indices based upon subjective and categorical criteria as proposed by Haro and Krueger (1988) for A. *rostrata* (Figure 4) and by Elie and Lecomte-Finiger (1982) for A. *anguilla* (Figure 5) while useful as a general measure of development they are difficult to relate to other important, continuous, variables such as measures of body size.

My research evaluates the potential usefulness of image analysis quantification of external pigmentation as a continuous variable, which potentially provides a more informative account of the change in external pigmentation during the migration. Although it is unclear how these developments affect recruitment, collecting data in the most informative way improves the ability to investigate unanswered questions.

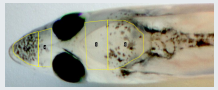
Materials and Methods

Elver collection and biological sampling:

1. Elvers were collected using Irish style elver boxes at East River, Chester, Nova Scotia.
2. Three times each week one hundred elvers were randomly sampled from the total catch and measured for length and weight.
3. Sedated elvers were placed, along with a stage micrometer (0.1mm), on an Epson digital scanner and scanned using the thumbnail 3200dpi 24-bit colour setting. Images were taken of the lateral and dorsal views of elvers.

Image analysis:

1. Length of the elvers contained in the scanned images were measured again using ImageJ and with reference to the stage micrometer.
2. Areas of interest were defined by landmarks on the elver and were outlined with the polygon selection tool. A distribution was created of the intensity values of each pixel. Statistics were calculated that were representative of the distribution.
3. Additional information collected from images included the presence and absence of brachial arches and food items in the gut. Pigment was reassessed using the Haro and Krueger (1988) and Elie and Lecomte-Finiger (1982) methods.



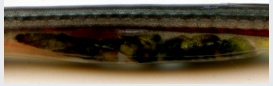


Figure 6. Map of East River Chester, NS

Objectives

1. Evaluate the potential usefulness of image analysis software, ImageJ, to quantify the development of elver pigmentation over the migration period as a continuous variable using pixel intensity.
2. Investigate the usefulness of ImageJ to estimate the total length of elvers.
3. Investigate how length and weight change over migration in relation to time and pixel intensity.
4. Observe the presence or absence of food items in the gut, as well as the presence or absence of brachial arches, both compared to pigment and time.
5. Compare the difference between how the Haro and Krueger (1988) and Elie and Lecomte-Finiger (1982) categorical stages of pigment change over time.

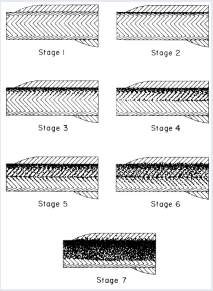


Figure 4. Haro and Krueger (1988) criteria

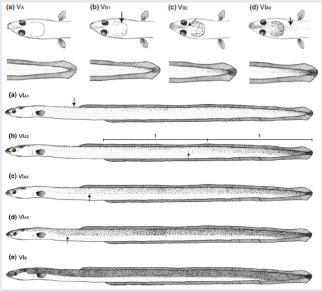


Figure 5. Elie and Lecomte-Finger (1982) criteria

Results

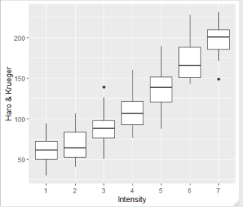


Figure 7. Pixel intensity in the area used for Haro and Krueger criteria exhibits a large amount of overlap between stages.

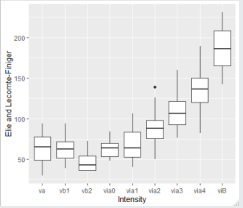


Figure 8. Pixel intensity in the area used for Haro and Krueger (1988) criteria exhibits a large amount of overlap between Elie and Lecomte-Finiger (1982) stages. While this is expected in stages va-viaO, stages via1-vB also exhibit large amounts of overlap.

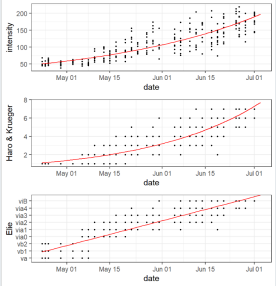


Figure 9. During the beginning of the run glass eels exhibit low variability in pigment. Variability increases over the run as pigmented elvers became more abundant.

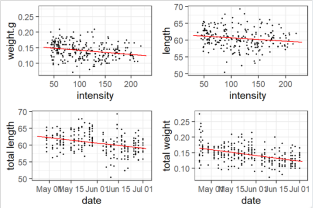


Figure 10: Throughout the run elver total length and weight decrease as intensity of external pigmentation increases.

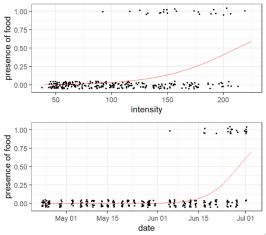


Figure 11. Food items are more likely to be observed as present in the gut later in the migration in highly pigmented elvers.

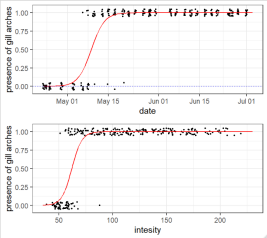


Figure 12: During the beginning of the run glass eel brachial arches are not pigmented but develop over time as external pigmentation increases.

Acknowledgements

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