Stock Assessment of Walleye (*Sander vitreus)* in Waterhen Lake, MB

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Abstract

Eco-certification of fisheries is becoming more prevalent as large retailers now insist on eco-certified seafood. A key step in this process is stock analysis to assess age structure, reproductive capacity and sex composition of the harvested population. The walleye fishery at Waterhen Lake is small scale which makes it ideal for an initial eco-certification assessment. Walleye caught on this lake in gill-nets were weighed and measured post-catch. Annular growth rings on opercular bones and a subsample of otoliths were examined to determine the age distribution of the fish. These ages were used to determine age at maturity and vulnerability of age classes to mesh sizes used by the fishery. Current harvest policy was evaluated to ensure that sexually immature fish are not being harvested before reaching reproductive age and it was found that the province's minimum mesh size of 3.75" (95 mm) may be sufficient for protecting pre-reproductive fish but analysis with a larger sample size is recommended. Back calculated lengths-at-age could be used to determine age and growth data from opercula in fisheries waste, but were found to underestimate due to Lee's Phenomenon and time of sampling. A stock assessment of Waterhen Lake walleye is an important element in the process of eco-certification of this fishery.

Acknowledgements

I would first like to thank my supervisor Dr. Scott Forbes without whom I would have very little fish knowledge and no answers to my endless questions. To my committee members – Nancy Loadman, who always makes me laugh, and Geoff Klein, who helped me survive a long day on Waterhen with a bum engine – thank you for your help throughout this project. To Brian McGregor, for help with the map and a ride home in the rain. Thanks also to Dr. Murray Wiegand, the Thesis Course coordinator, who was an invaluable resource this year.

Big thanks to my family and friends who have put up with me talking about fish all year, especially to my Uncle Den who got me started in the fish business, my lab mate Tim, who can find the humour in anything (even boiling fish heads), mon chum, qui est toujours là quand j’en ai besoin, and to my iPod, without which I would not have been able to survive all the hours in the lab.

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Introduction

Overfishing is widely recognized as a serious challenge for global fisheries, both marine and freshwater (Pauly *et al.*, 2002; Jackson, 2001), and there is growing consumer awareness that many fisheries are managed on an unsustainable basis. Eco-certification is a process whereby an independent agency evaluates harvesting practices on the basis of ecological, economic and sociological sustainability (Gulbrandson, 2005). In fisheries, the Marine Stewardship Council (MSC) is widely considered as the gold standard for eco-labelling. It has developed a rigorous set of standards for evaluating fisheries to ensure that wild-capture fisheries are ecologically sustainable and well-managed, and to implement sustainable fishing practices on fisheries to ensure the fish populations and the ecosystems on which they depend remain healthy and productive (MSC website) .

Eco-certification is becoming increasingly necessary as major fish buyers, particularly in Europe and North America, are demanding fish which come from eco-certified populations (Gulbrandson, 2005). This poses a major challenge for commercial fisheries in Manitoba where currently no fish populations are MSC certified.

MSC eco-certification requires detailed biological information about the structure of the harvested population, including the age, size and sex structure of the stock (MSC website). This baseline information is required to assess whether the harvest is conducted in an ecologically sustainable manner.

In Manitoba, the most important species in the commercial fishery is walleye (*Sander vitreus)*. The fishery comprises 39% of total production by weight and 70% of the average landed value of the total catch for Manitoba (Manitoba Water Stewardship Fisheries Branch, 2010). Walleye are one of the two key harvested species (the other being the northern pike, *Esox* *lucius*) on Waterhen Lake, MB. This is a smaller fishery that is being considered for MSC eco-certification (Geoff Klein, Manitoba Fisheries Branch, pers, comm.).

There are a variety of structures that are used in aging fish including the otoliths, opercular bones, scales and dorsal spines. Otoliths are usually considered the most valid aging structures (Erickson, 1983; Kocovsky and Carline, 2000). Opercular bones however are often used by fisheries biologists for age analyses (Babaluk *et al.*, 1993; Cooley and Franzin, 1995) and usually produce age estimates similar to those produced with otoliths. In this study, opercular bones were used which are more easily accessed and exhibit clear growth patterns, with a subsample of otoliths for validation.

Walleye Life History

Walleye (*Sander vitreus*), also known as pickerel, are part of the subfamily Luciopercinae of the Family Percidae. Along with sauger, members of this subfamily are slender and round in cross section with strong, fang-like teeth and sharp dorsal fin rays. The walleye is widespread in Manitoba, with its distribution covering almost the entire province (Stewart and Watkinson, 2004). This species is typically found in deep, non-turbid waters such as Manitoba's Canadian Shield lakes and rivers. Walleye (along with northern pike, *Esox lucius*) are top fish predators in Manitoba's freshwater ecosystems. However, northern pike are higher on the food chain than walleye and tend to prey on walleye over much of their range. It may also be in direct competition with walleye for resources in shallow waters in the north (Scott and Crossman, 1998). Walleye change their feeding habits as they increase in size. Walleye feed on zooplankton until they reach around 30-50 mm total length (TL) when they switch to feeding on macroinvertebrates. Near 60 mm TL they become piscivorous (Hoxmeier, 2006; Preigel, 1969;

Mathias and Li, 1982), eating other fish including juvenile freshwater drum, white bass and yellow perch (Isermann, 2007). Juvenile walleyes are also subject to predation by larger species such as the northern pike and channel catfish (Stewart and Watkinson, 2004).

Walleye spawn in water temperatures of 4°C shortly after the ice breaks up, the timing of which can vary from mid- to late April to May (Stewart and Watkinson, 2004), depending on temperature and latitude (Scott and Crossman, 1998). Mature walleye may travel south into warmer waters to spawn, where the females release their eggs over a rocky substrate and mature males fertilize the eggs with their milt, and leave the fertilized eggs to develop (Stewart and Watkinson, 2004). The eggs will then clump to the substrate to increase hatching success by preventing eggs from falling into the silt.

Walleye grow fast in the first few years of life. Growth rates decrease after maturation of the fish as the bulk of energy has shifted to reproduction as opposed to somatic growth (von Bertalanffy, 1938). However, changes in reproductive strategies of a species can occur in particular environmental conditions. For example, limited resources will cause a delay in reproduction until somatic growth is nearly complete. Populations of fish that have been exploited are the opposite, as they have more energy available to them due to decreased population numbers and thus grow faster. These fish reach maturity earlier and are able to then focus on reproduction at an earlier age (Craig *et al*., 1995; Trippell, 1995).

Age-at-maturity is defined as the age at which the majority of fish are determined to be sexually mature based on external examination of the gonads (Babaluk *et al.*, 1993; Madenjian *et al*., 1996). In walleyes, this depends on a whole host of environmental factors including temperature, prey availability and predation. Fishing pressure is also found accelerate the onset of maturity as a response to high fishing mortality rates (Scott and Crossman, 1998).

Project Goals

The objective of my thesis research was to assess the age, size and sex structure of walleye in Waterhen Lake. Such information would potentially be valuable in the process of eco-certification. At present, there is only one MSC eco-certified freshwater fishery in the world: pikeperch (*Sander lucioperca)* in Lake Hjälmaren, Sweden (Tuene and Hough, 2007). I have used the eco-certification of this fishery as a template for my own work.

The specific objectives of this study were five-fold:

1. To assess the age structure of male and female walleye in Waterhen Lake;
2. To assess the age of maturity in male and female walleye in Waterhen Lake;
3. To assess the vulnerability of different age/size classes of walleye to gill-nets of different mesh size;
4. To assess the current harvest policy with respect to mesh size restrictions – i.e., are walleye being harvested before they reach maturity; and
5. To develop statistical models of the relationship between the dimensions of opercular bones and the length and mass of Waterhen Lake walleye. This will allow future workers to collect discarded heads from the commercial fishery to assess the age and size structure of fish in the harvest.

Methods

Study Area

Waterhen Lake is located 270 km Northwest of Winnipeg (52.08^{o}N, 99.58^{o}W, see Appendix A for map), with Lake Winnipegosis to the north and Lake Manitoba to the south, connecting to Waterhen Lake via the Waterhen River. The main sources of income for people on Waterhen Lake include commercial fishing, trapping and livestock production. Commercial fishing is important to the local economy in the Interlake and Northern Manitoba. Aside from the three major producing lakes in Manitoba (Lake Winnipeg, Lake Manitoba and Lake Winnipegosis) there are 295 other lakes listed in the provincial commercial harvest schedule by Manitoba Water Stewardship (2010). There was a fish packing station located in Skownan, on the Eastern side of Waterhen Lake. This lake, along with others categorized as "Other Lakes" in the Manitoba Water Stewardship *Profile of Manitoba Fisheries*, contributes 18.2% of yearly production of fish in Manitoba. Commercial fishing on Waterhen Lake is small scale, consisting of just over 20 fishers, harvesting mainly walleye and pike. This is a smaller fishery that is being considered for MSC eco-certification (Geoff Klein, Manitoba Fisheries Branch, pers. comm.).

Sample collection

Samples of fish were collected from September 11 to September 23, 2011 in routine annual test nets set by provincial fisheries biologists. Test netting consists of overnight sets of gangs of gill-net mesh from 2 to 5” in ½” intervals. The captured walleye were weighed to the nearest 5 g and measured for total length (from the snout to the extreme tip of the longest caudal lobe with the lobes compressed) to the nearest mm. The sex of the fish and whether it was sexually mature, determined by external examination of the gonads, were recorded. The date and location of capture were also recorded, as was the mesh size from which the fish was captured (for a subsample of walleye). Heads were removed from the captured walleye, labelled with an identification number, and delivered to the University of Winnipeg.

Opercular Extraction, Processing and Age Determination

The right and left opercular bones were removed from the fish by prying them off the head with a blunt probe and ripping off as close to the skull as possible, being careful not to crack the opercular bone. For difficult specimens, the head was placed in hot water to make the bone easier to remove. Once removed from the fish, the opercular bones were placed in boiling water to loosen the skin, which was then completely rubbed off with paper towel. The opercular bones were rinsed and allowed to dry. Once dry, the opercular bones were viewed either with the naked eye or under a magnifying glass with a contrasting background which allowed the annuli to be read easily. Age was determined by counting the annuli for both the left and right opercular bones twice, counting on non-consecutive days to decrease reader bias. A subsample of 50 opercular bones was counted by a second reader to increased validity.

Counting annuli to determine fish age is much like reading the age of a tree from the rings. Two rings are laid down per year determined by the environmental conditions. A clear ring (composed mostly of protein) forms in the warm summer months due to increased feeding and fish growth. A dark ring (composed of minerals) which appears in winter when feeding and growth are minimal (Scott and Crossman, 1998). This same effect is seen in otoliths.

Otolith Extraction, Processing and Age Determination

Otoliths are the inner ear bones of fish which aid in balance and hearing. There are three otoliths on each side of the skull (saggita, asteriscus, and lapillus). Of these, the saggital otolith was used as an age structure as it is the largest of the otoliths and, like the opercular bones, has seasonal growth rings (annuli) that are deposited in the bone and read as yearly markers. The pair of saggital otoliths were dissected out of the head of a subsample of 20 walleye. One was kept and the other was sent to Manitoba Fisheries. Extraction of the otoliths was via a cut through the anterior end of the isthmus ahead of the gill arch junctions thus exposing the posterior part of the chamber where the saggital otoliths are housed. If frozen within, warm water was applied until the otoliths could be extracted without breaking them. Otoliths were then rinsed with water and allowed to dry. The nucleus of each otolith was marked with an ink dot and allowed to cure completely for about a week. Epoxy resin was prepared by mixing 2 parts Cold Cure hardener with 1 part Cold Cure resin. Individual ice cube tray compartments were labeled with corresponding specimen numbers. The otolith was placed in its ice cube tray compartment, immersed in epoxy and allowed to cure for at least a week. When the blocks cured, a thin section was taken by making two consecutive transverse cuts, ~0.5 mm apart, with a low RPM Buehler Isomet saw containing a diamond chip blade. The two cuts were made on either side of the marked nucleus so as to contain the nucleus and all annular growth rings. The thin sections were then viewed under a dissecting microscope using transmitted light in order to count the annuli.

Data Analysis

Data were entered into a Microsoft Excel spreadsheet and included sample number, date, location, set number, mesh size, weight, total length, maturity status, sex, and age as determined by both opercular bones and otoliths. Regression analyses including total length at age, total length at weight, maturity at age, maturity at length and mesh size at age/length/weight were constructed in Microsoft Office Excel. The deviation between opercula estimated age and otolith estimated age, as well as the deviation of estimated ages between readers was calculated.

Measurements of the annuli on opercular bones were used to back-calculate length-at-age for walleye in my sample – e.g., how long was a 6-year-old walleye when it was 5, 4, 3, 2 and 1 year(s) old? As total length is linearly related to opercular length, length-at-age is calculated as a simple fraction of current total length. For example, if the distance to the annulus for age "x" is 2/3 of the total operculum length, the length of the fish at age "x" was 2/3 of its current total length.

Opercular bones were scanned using a CanoScan LiDE 210 flatbed scanner and the image files stored in the computer based on their sample number. Left opercular bones were used wherever possible, and right bones were used where the left were broken or incomplete. Measurements using the software program ImageJ started at the focus of the bone and were measured to the outer edge of the annuli as well as distance between the annuli along an axis which roughly bisected the opercula into dorsal and ventral halves following the method of Le Cren (1947). This axis was chosen in contrast to the method described in Cooley and Franzin (1995) where the axis used was closest to the ventral spinous process of the operculum. When reading manually (as opposed to using an Image Analysis Program which detects variances in luminescence on the bone) the primary annuli are more obvious and so were read here at a perpendicular to an axis bisecting the bone in two roughly equal halves (Figure 1). All measurements of annuli were determined manually, entered into an Excel spreadsheet, and compared to age and length.

Actual length of the operculum was measured in 20 fish to the nearest 0.1mm with a caliper and compared to those lengths in the arbitrary units used on the ImageJ software program. The mean of these measurements was used as a conversion factor between ImageJ units and actual length of the opercula. Back-calculation was used to determine the length-at-age for these fish and compared to mean empirical length-at-age for the sample from Waterhen Lake using linear regression.

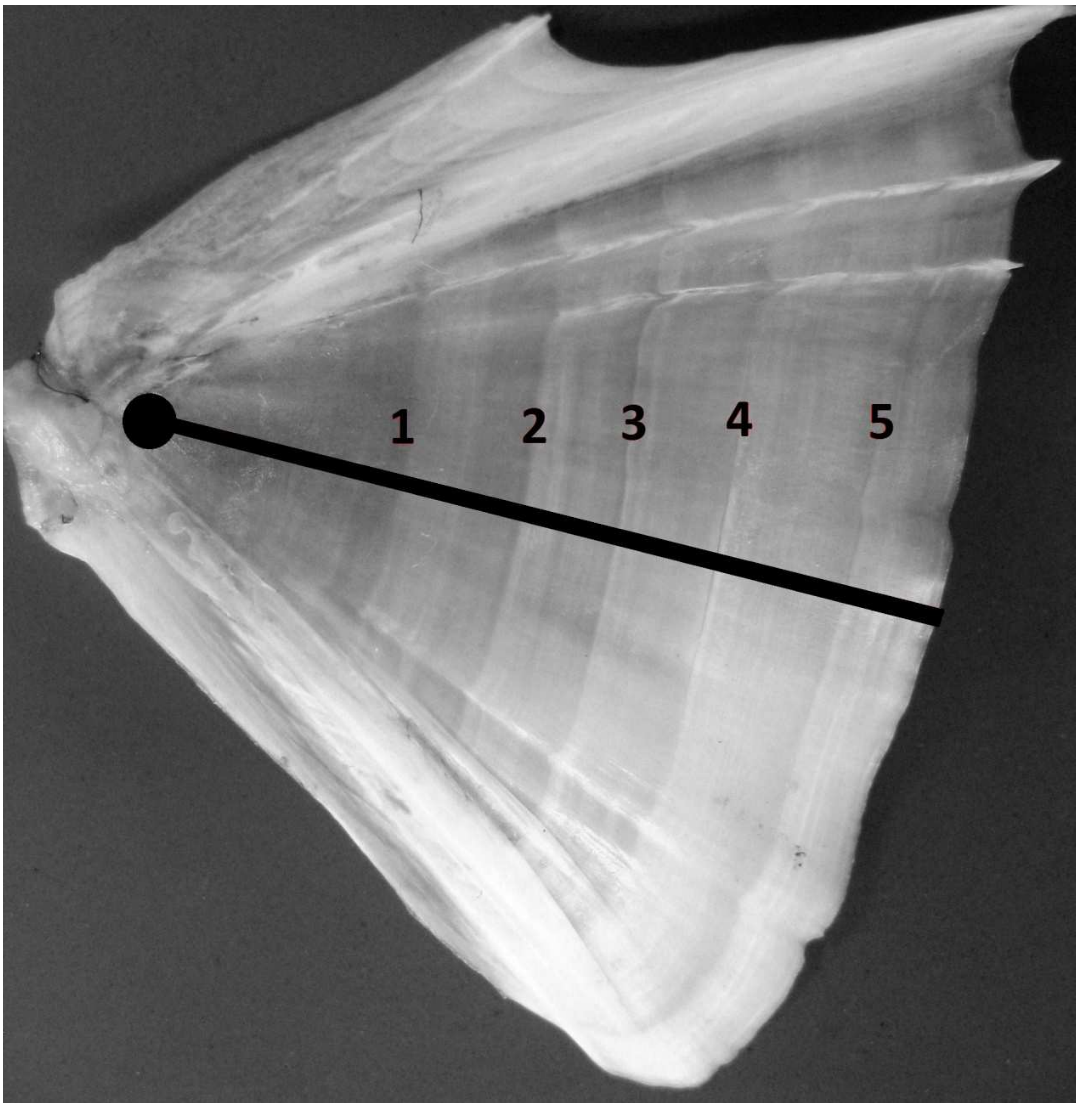

Figure 1. Left walleye operculum showing origin (dot) and bisecting axis (line) along which measurements for back-calculations of length-at-age were made.

Results

Age and Growth

161 walleye from Waterhen Lake were aged using opercular bones. Age ranged from 0 to 10 years, with a mean and median of 3 years (Figure 2). Disagreement of ages between the two readers was minimal, almost always resulting in an agreement when reviewing the age structure in question together and deciding upon a final age. Age estimates based on otoliths were compared with age estimates from opercular bones using linear regression. There was a strong linear relationship (Figure 3, $\beta = 0.998$, $R^2 = 0.953$, $F_{1,18} = 366.6$, $P < 0.001$) indicating that ages estimated from opercular bones corresponded closely with ages from otoliths.

The annuli on both the opercular bones and otoliths of walleye are fairly easily read (Figures 4 and 5) with the exception of the first annulus which is sometimes not as clear. Growth comparing total length of the fish against age is described by the von Bertalanffy growth model along with its parameters. Female walleye (Figure 6, Table 1) reach an ultimate length of 696.9 mm where male walleye (Figure 7, Table 2) are smaller with an ultimate length of 522.70 mm. The total lengths ranged from 158 mm in a young-of-the-year walleye to 679 mm in a 10-year-old walleye. The logarithmic relationship between mass and length of Waterhen Lake walleye was nearly perfectly isometric (Figure 8, Mass (g) = 5×10^{-6}(TL (mm))$^{3.107}$, $R^2 = 0.997$, $F_{1,172} = 27416$, $P < 0.001$).

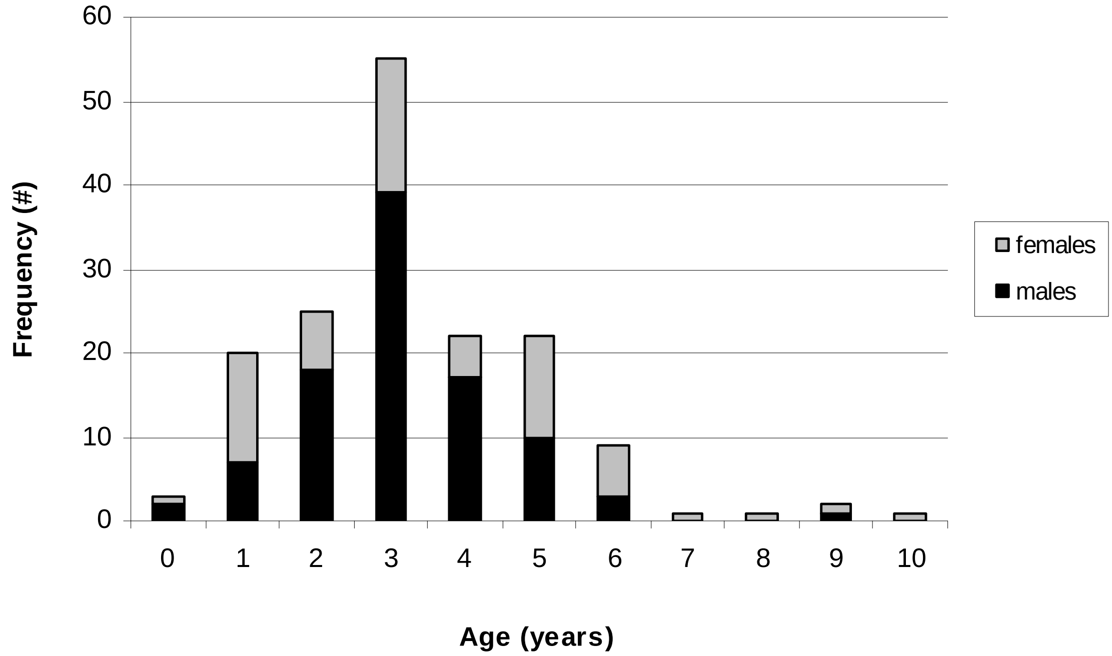

Figure 2. Age frequency distribution of walleyes caught in experimental gill nets (n=161) from Waterhen Lake, MB.

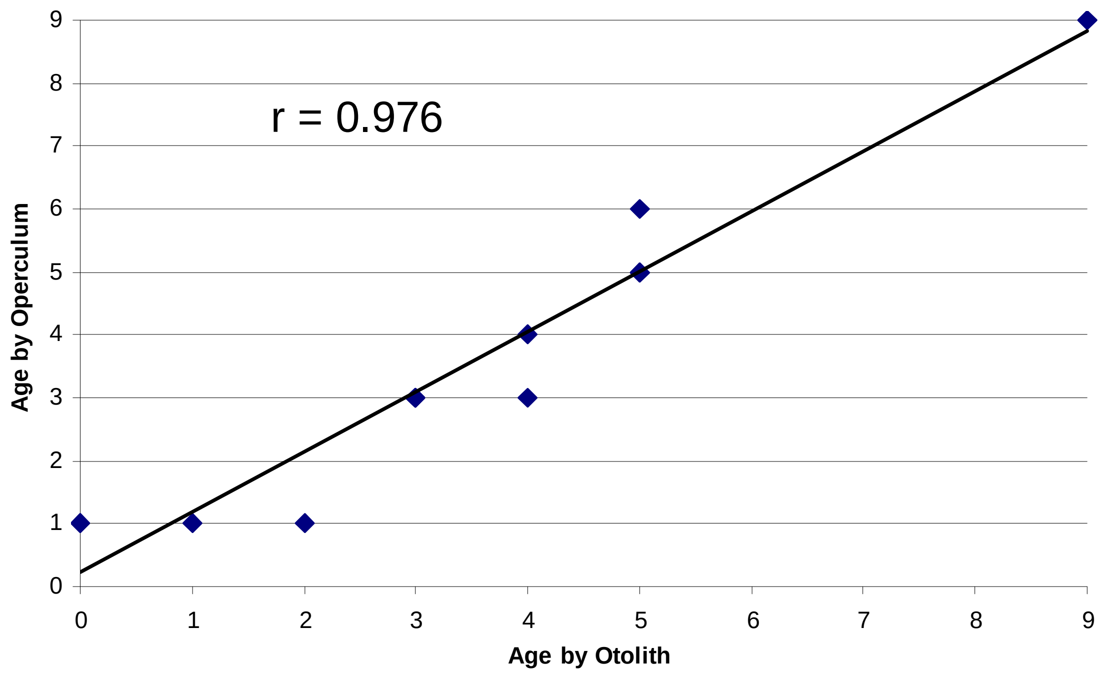

Figure 3. Age estimate comparison by use of otoliths and opercular bones (n=20). A correlation of 1.00 would represent a perfect match between both aging techniques.

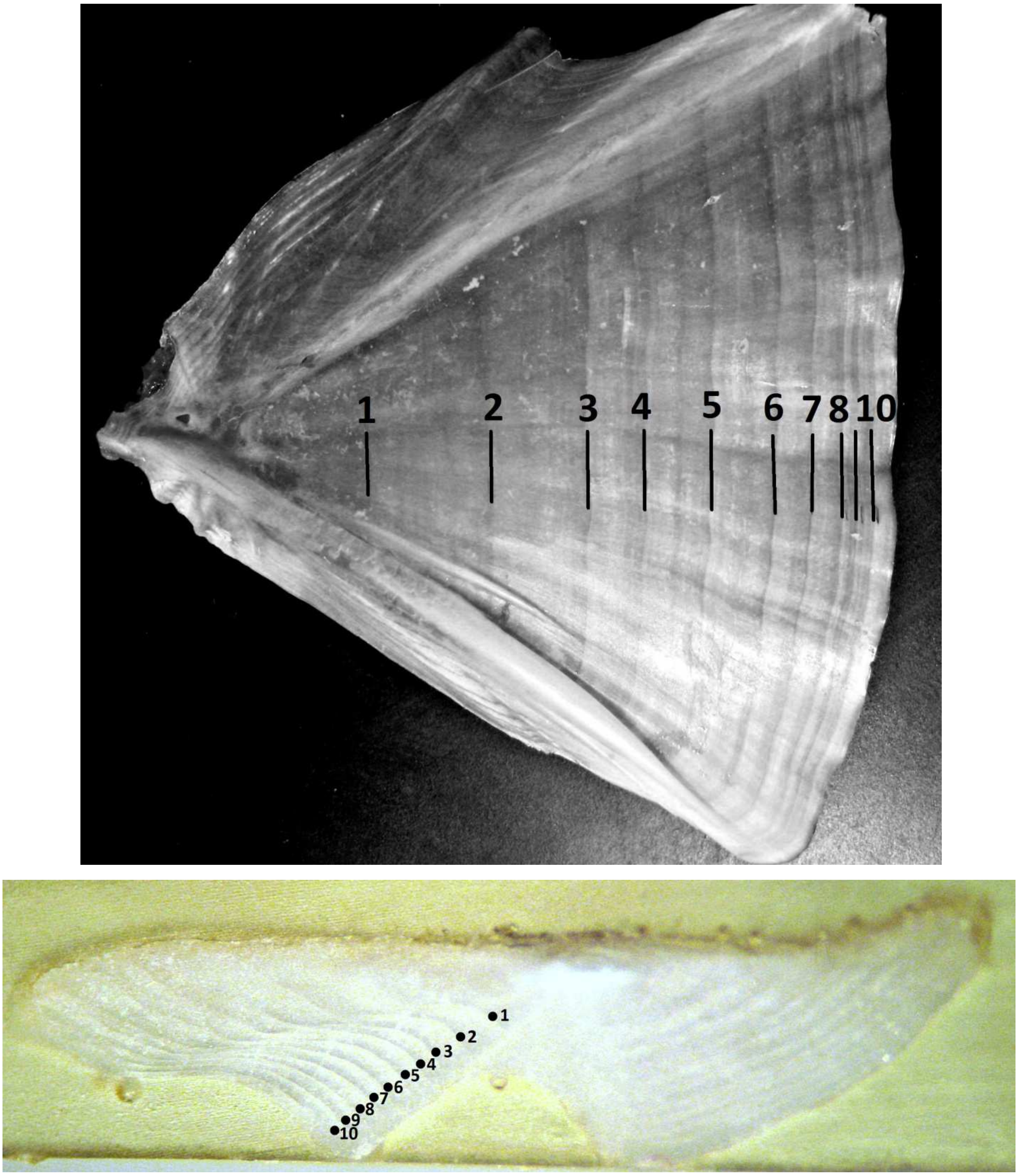

Figure 4. Operculum (above) and otolith section (below) from the same 10 year old fish. Both aging structures gave an age of 10 years. Annuli are marked and numbered.

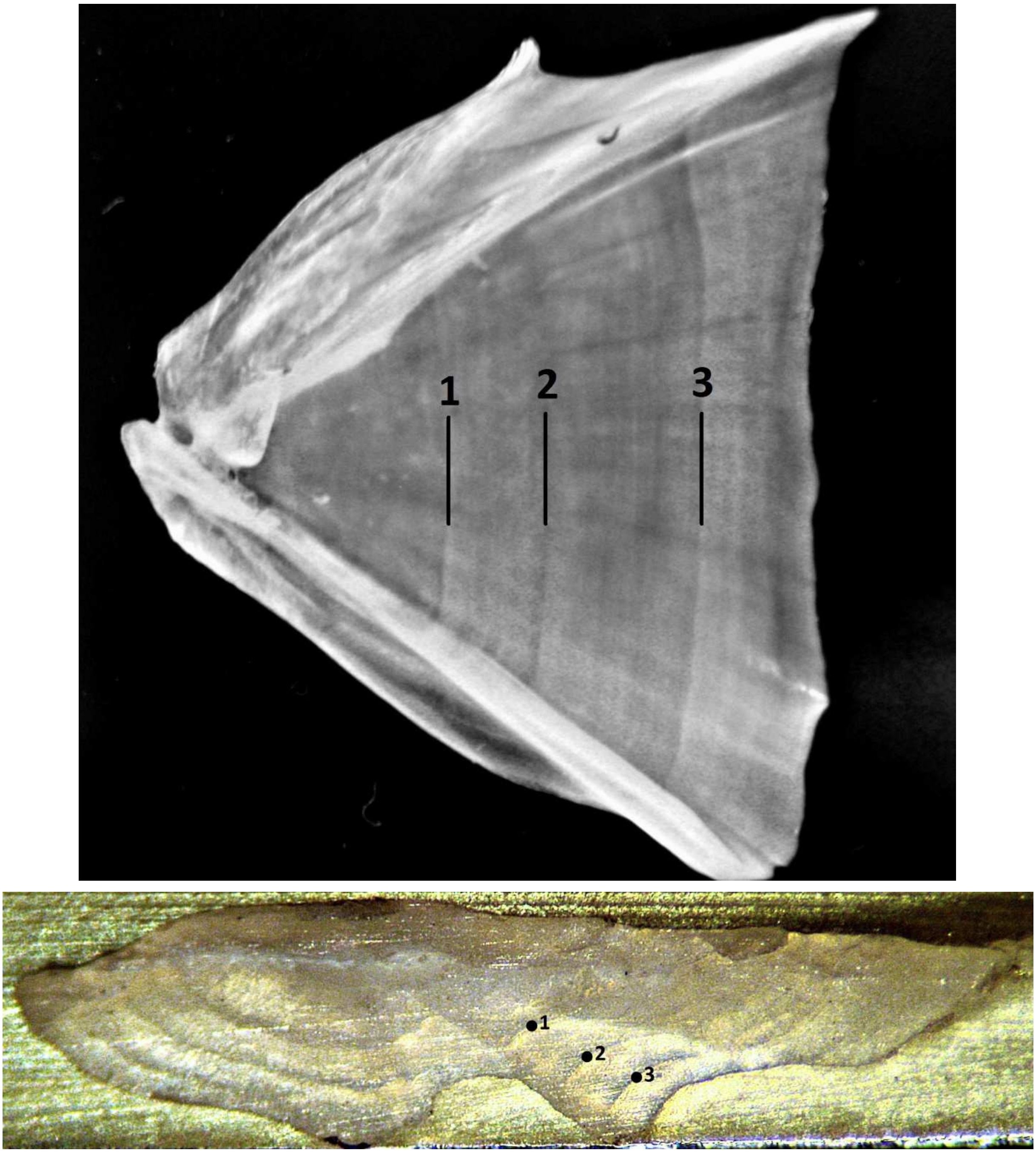

Figure 5. Operculum (above) and otolith section (below) from the same 3 year old fish. Both aging structures gave an age of 3 years. Annuli are marked and numbered.

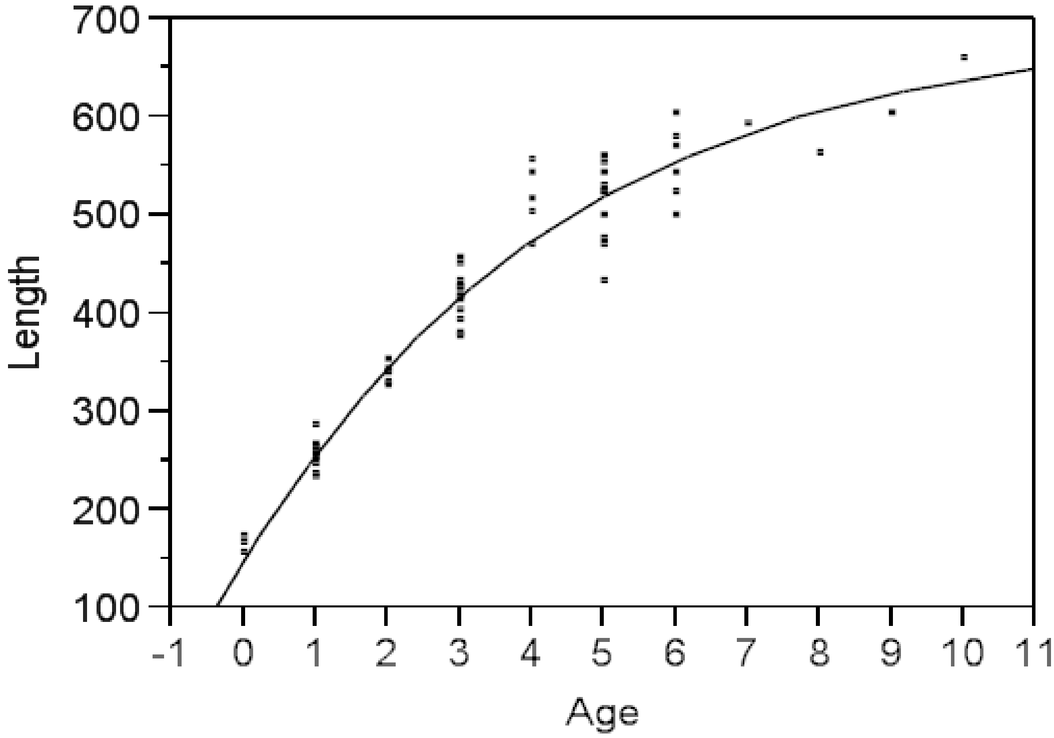

Figure 6. von Bertalanffy curve of total length (mm) at age as determined by opercular bones of female walleye (n=64) from Waterhen Lake, MB.

Table 1. Values for von Bertalanffy growth parameters for fitting growth curves of female walleye from Waterhen Lake, MB.

FEMALES		
L∞	**K**	**t_0**
696.59	0.244	-1.242

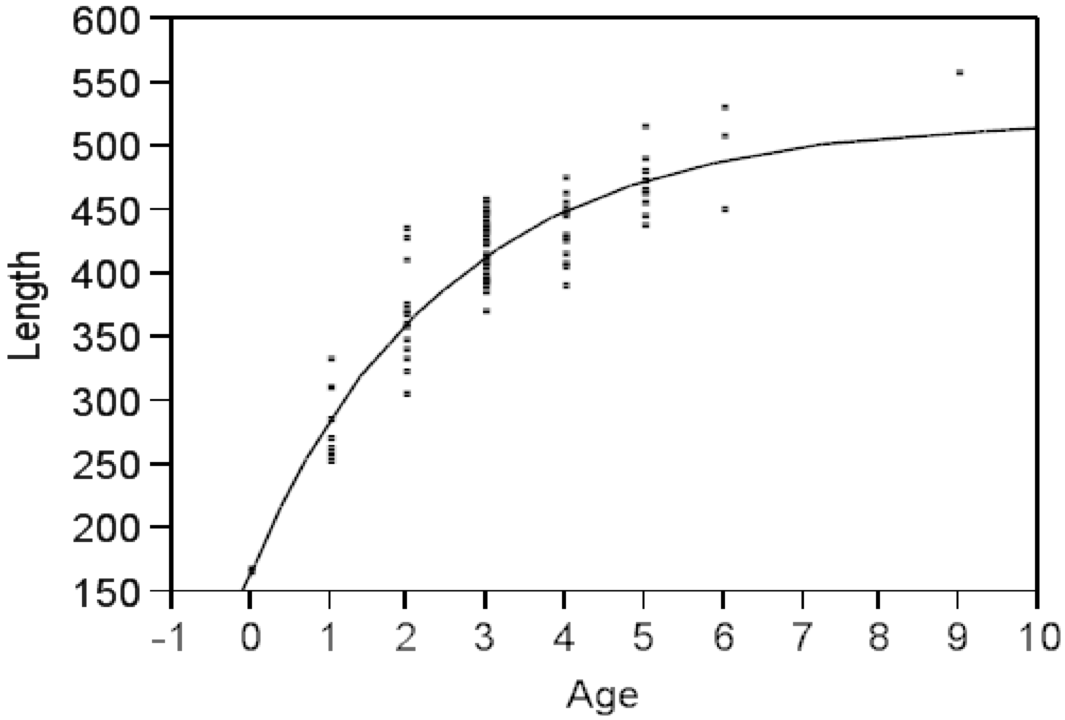

Figure 7. von Bertalanffy curve of total length (mm) at age as determined by opercular bones of male walleye (n=97) from Waterhen Lake, MB.

Table 2. Values for von Bertalanffy growth parameters for fitting growth curves of male walleye from Waterhen Lake, MB.

MALES		
L∞	**K**	**t_0**
522.7	0.395	-0.982

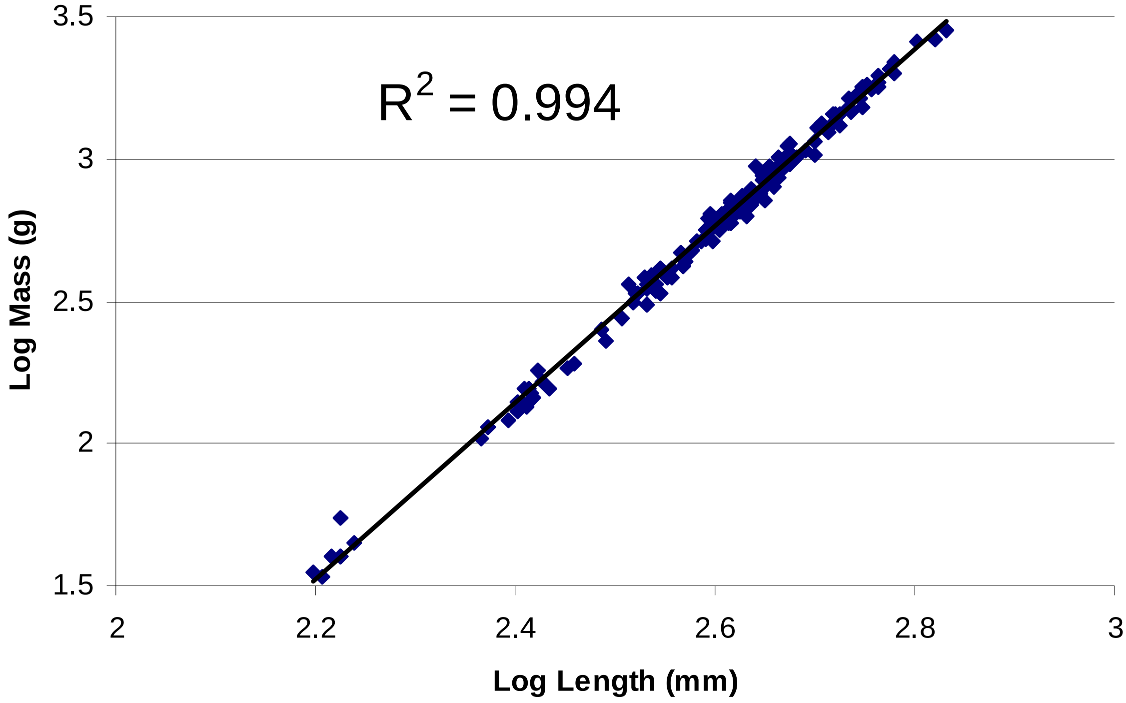

Figure 8. Weight (g) at total length (mm) of walleye (n=161) from Waterhen Lake, MB.

Age at Maturity

Age at maturity differed by sex in Waterhen Lake walleye. Most (92.3%) males were mature at age three, whereas no females were mature at age three. All four-year-old females (100%) were mature (Table 3). These data suggest that male walleye are likely to mature between the ages of 1 to 4 years, therefore first spawning the following spring (when temperatures increase) at 2-5 years. Female walleye will reach maturity between 4 to 6 years and first spawn from 5-7 years. A logistic regression (Figure 9) shows this range of maturation for both male and female walleye. Both of these regression lines were highly significant (Table 4) which shows that males do in fact mature earlier than females.

Table 3. Maturity schedule for walleye from Waterhen Lake, MB (Numbers of mature male and female walleye over total number caught, percentage mature fish at each age in parentheses).

	Age Class										
sex	**0**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**
M	0/2	1/6	6/18	36/39	17/17	10/10	3/3	-	-	1/1	-
	(0.0)	(16.7)	(33.3)	(92.3)	(100.0)	(100.0)	(100.0)			(100.0)	
F	0/1	0/13	0/7	0/16	5/5	11/12	6/6	1/1	1/1	1/1	1/1
	(0.0)	(0.0)	(0.0)	(0.0)	(100.0)	(91.7)	(100.0)	(100.0)	(100.0)	(100.0)	(100.0)

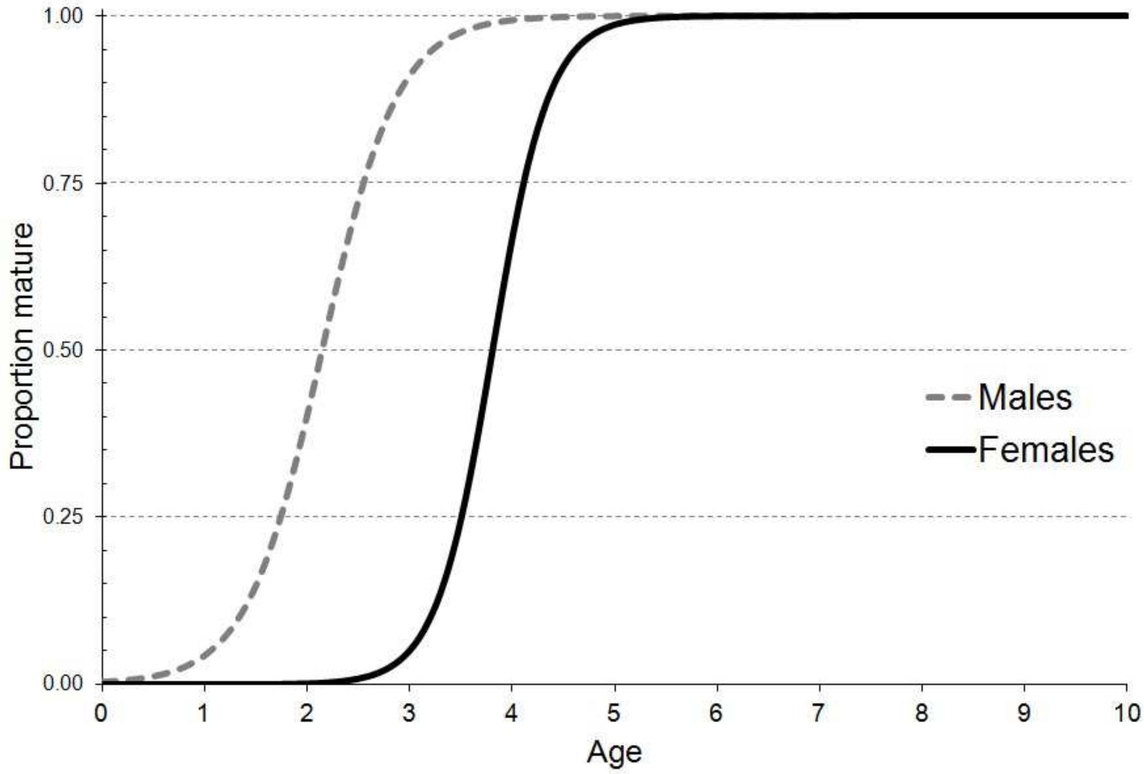

Figure 9. Proportion of mature male and female walleye in relation to age (years) from Waterhen Lake, MB.

Table 4. Logistic regression results for age at maturity of male and female walleye from Waterhen Lake, MB.

	Constant	**β**	**Wald statistic**	**df**	**p**	**Cox & Snell R^2**
Males	-5.829	2.719	20.173	1	<0.001	0.431
Females	-13.78	3.619	10.315	1	0.001	0.674

Vulnerability to Mesh Sizes

Data on the mesh size at which walleye were captured was not personally collected. Instead, data gathered by Geoff Klein (Manitoba Fisheries Branch) in 2010 and 2011 was used to assess the vulnerability of different age and size classes of walleye on Waterhen Lake to gill nets at different mesh size. Stretched mesh sizes ranged from 2” to 5” (or 51-127 mm) in 0.5” intervals. A variety of fish sizes were caught in each mesh size but the general trend showed the fish with a greater total length being caught in larger mesh sizes. A linear regression analysis showed a slightly positive linear relationship between the length of fish captured and mesh size (Figure 9, Length at capture (mm) = 2.38 × Mesh size (mm) + 254.1, $R^2 = 0.463$, $F_{1,143} = 37.1$, $P < 0.001$). That is, younger fish were caught in smaller meshes while older fish were caught in larger meshes. The largest mesh size (127 mm or 5”) caught a fish with a total length that would estimate its age at ~7-8 years old where a mesh size of 102 mm (4”) caught both an estimated 3-4 year old and an estimated 9-10 year old walleye (Table 4). Percent immature fish caught in the gill nets decreased with increasing mesh size (Table 5), indicating a low vulnerability of immature fish to the fishery.

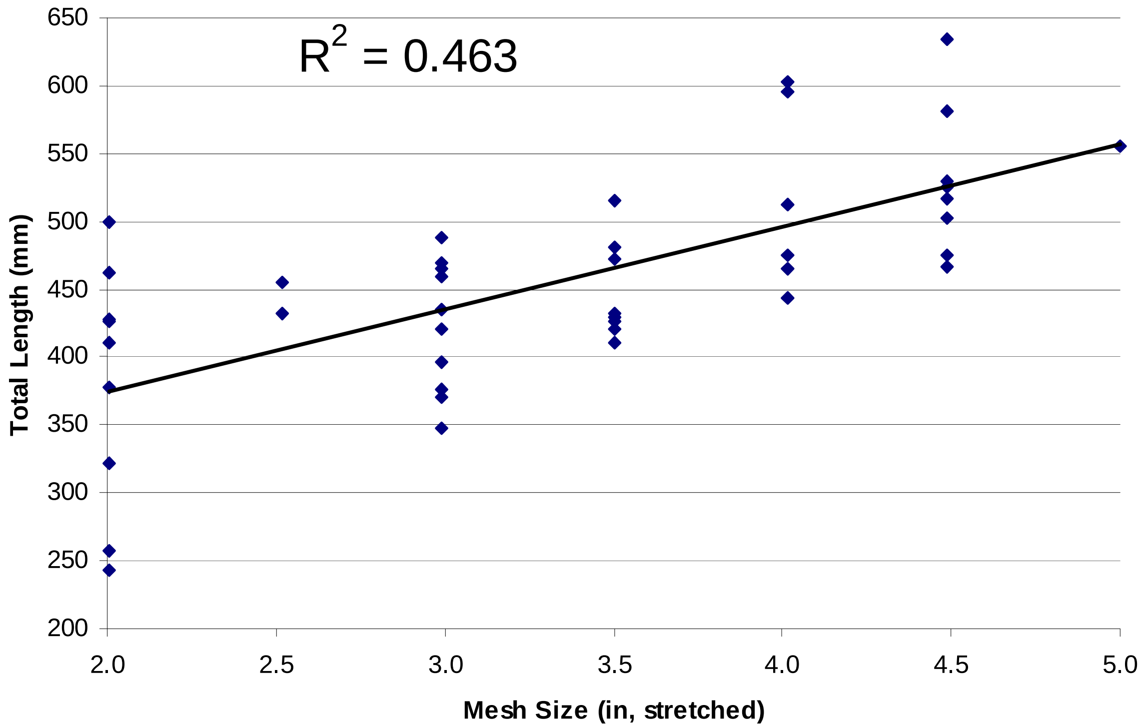

Figure 10. Total length (mm) of walleye caught in differing mesh sizes (mm) of gill nets on Waterhen Lake, MB (2010-2011 sampling years).

Table 5. Recruitment of different aged walleye to varying mesh sizes used by Provincial Fisheries Biologists in routine test netting on Waterhen Lake, MB.

		Estimated Age (years)								
MESH SIZE (in)	**n**	***1-2***	***2-3***	***3-4***	***4-5***	***5-6***	***6-7***	***7-8***	***8-9***	***9-10***
2.0	**9**	3	1	3	2	-	-	-	-	-
2.5	**2**	-	-	2	-	-	-	-	-	-
3.0	**11**	1	2	5	3	-	-	-	-	-
3.5	**8**	-	-	5	2	1	-	-	-	-
4.0	**6**	-	-	1	2	1	-	-	1	1
4.5	**8**	-	-	-	2	1	3	1	-	1
5.0	**1**	-	-	-	-	-	-	1	-	-

Table 6. Percent mature fish caught in varying mesh sizes used by Provincial Fisheries Biologists in routine test netting on Waterhen Lake, MB.

MESH SIZE (in)	**2.0**	**2.5**	**3.0**	**3.5**	**4.0**	**4.5**	**5.0**
n	**9**	**2**	**11**	**8**	**6**	**8**	**1**
% immature	66.7	50.0	36.4	12.5	16.7	12.5	0.0
% mature	33.3	50.0	63.6	87.5	83.3	87.5	100.0

Back-calculation of Length-at-Age

The relationship between length of the operculum and total length of the fish was constructed (Figure 10, R^2=0.923) and was found to be a strong linear relationship. A reduced major axis regression was also performed to correct for random error in both the *x* and *y* axes (operculum length [*x*] and total length [*y*]) and validates the use of the opercular bones as back-calculation structures.

Table 6 summarizes back-calculated lengths-at-age for walleye in my sample. Mean empirical lengths at capture were greater than mean back-calculated lengths for almost all age classes, especially those in the 1-7 year age classes. Back-calculated lengths-at-age agreed reasonably with mean empirical lengths at capture in older age classes (8-10 years).

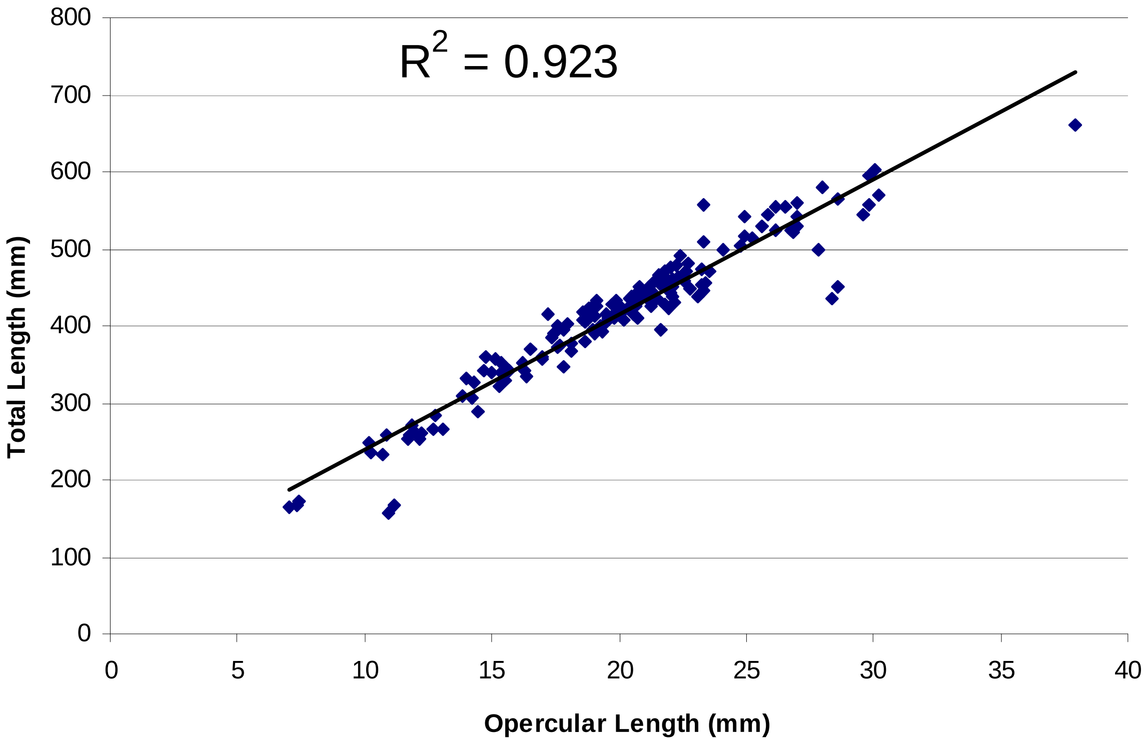

Figure 11. Body – opercular relationship for walleye from Waterhen Lake, MB.

Table 7. Mean back-calculated total lengths-at-age for walleye from Lake Waterhen.

		annulus no.										
age	n	1	2	3	4	5	6	7	8	9	10	edge
0	3											168.3
1	20	**145.6**										255.7
		28.9										
2	25	**145.9**	**248.6**									355.4
		30.4	35.5									
3	55	**139.0**	**230.5**	**327.1**								417.7
		17.5	18.8	26.1								
4	22	**153.1**	**233.2**	**312.8**	**387.6**							450.5
		29.5	31.8	33.7	33.4							
5	22	**165.3**	**241.6**	**316.6**	**383.9**	**441.5**						498.7
		21.1	29.9	31.7	32.8	36.4						
6	9	**151.2**	**237.6**	**323.1**	**386.4**	**437.2**	**485.1**					534.0
		28.2	22.7	32.5	34.9	41.8	45.9					
7	1	**136.9**	**211.3**	**302.6**	**370.2**	**434.5**	**502.1**	**562.9**				595.0
		-	-	-	-	-	-	-				
8	1	**131.8**	**187.6**	**261.3**	**319.4**	**390.8**	**431.2**	**480.1**	**504.7**			565.0
		-	-	-	-	-	-	-	-			
9	2	**153.7**	**233.5**	**296.8**	**350.7**	**418.7**	**470.1**	**506.9**	**529.8**	**555.7**		580.0
		29.1	31.4	25.1	38.6	51.1	49.0	32.1	31.7	26.5		
10	1	**123.9**	**183.8**	**260.7**	**334.2**	**404.8**	**477.7**	**541.0**	**574.6**	**601.0**	**625.5**	661.0
		-	-	-	-	-	-	-	-	-	-	
TOTAL	**161**											
mean back-calculated lengths (mm)		144.7	223.1	300.1	361.8	421.3	473.2	522.7	536.3	578.3	625.5	
annual growth increment		78.4	77.0	61.7	59.5	52.0	49.5	13.6	42.0	47.2		
mean empirical length at capture (mm)		258.0	357.9	418.3	456.0	497.2	534.0	595.0	565.0	580.0	661.0	

Discussion

My overall objective was to describe key life history characteristics of walleye in Waterhen Lake, MB in relation to exploitation in commercial fisheries in anticipation of assessment for eco-certification of the fishery. The starting point is to assess age and growth of the fish.

Age and Growth

Annular growth increments were reasonably easy to identify on both opercular bones and otoliths from walleye (Figures 4 and 5). In some cases of older fish (>6 years), the first and second annuli were not as apparent but could be inferred by the relative position of those annuli on a sample where they were more visible. Annuli crowding is usually seen in the oldest annuli towards the edge of the aging structure of older fish. Since the oldest in this sample was merely 10 years old annuli crowding did not pose a problem.

According to the Minnesota Department of Natural Resources website, a 27-year old Walleye was found in Lake of the Woods, a lake which is part of Manitoba, Ontario and Minnesota, in a routine sample taken in 2005. This walleye was 724 mm in length (28.5") which was not much larger than the total length of the oldest walleye found in this sample (661 mm at 10 years old). This shows that growth rates for walleye decrease with increasing age since the 12 year difference between the fish resulted in a mere 61 mm length difference. Looking at growth increments between years (Table 6) it is evident that growth rates are higher at younger ages and slow down after maturity is reached. However, the kinds of lengths seen at Lake of the Woods may not have been seen in Waterhen Lake because the walleye simply do not grow that large. In a comparison of growth and reproduction of walleye from lakes of different latitudes in

Manitoba, Craig *et al*. (1995) showed the walleye in the northernmost lake grew to smaller asymptotic sizes and at a slower rate. Growth rate for walleyes is rapid during the summer and is virtually non-existent in the cold winter months (Wolfert, 1977). Since northern lakes have a longer winter season than their southern counterparts, often with snowfall beginning in October and spring melt not occurring until late-April to May, their summer growing season is often cut short.

Age at Maturity

Age at maturity is an important parameter of fish life history and is inversely related to growth rate meaning when the emphasis is put on growth, maturation is temporarily less important. Due to this, age at maturity is closely correlated with temperature (Beverton, 1987) since faster growth rate (which is related to temperature) would cause earlier maturation (Craig *et al*., 1995). Walleye mature later the further north they live (Colby *et al*., 1979) because at higher temperatures, there is a longer period where prey is available and thus the growing period is extended. Northern lakes have a shorter growing period as the temperature is much lower, so reaching the minimum length-at-age for maturation takes longer.

Age at maturity for walleye is variable among stocks of differing geographical locations (Jensen, 1991), varying from 1-6 years for males and 2.3-7 years for females (Babaluk *et al*., 1993; DFO, 2010; Morgan *et al*., 2003; Sass and Kitchell, 2005; Wang *et al*., 2009). According to Scott and Crossman (1998) the general age of maturity for male walleyes is 2-4 years, or over 11" in length and for females is 3-6 years of age and 14-17" (356-432 mm).

Thus the walleye in Waterhen Lake, which were found to mature at 1-4 years for males and 4-6 years for females (Table 3), with age at first spawn occurring from 2-5 years in males and 5-7 years, have somewhat later maturation schedules compared to other lakes. The differences in age at maturity among lakes are caused by a number of factors like water temperature, latitude, resource availability and rate of exploitation. Latitude has been shown to strongly influence age at maturity, with populations further north maturing later, proven specifically for walleye in three Manitoba lakes by Craig *et al.* (1995). Fewer warm days because of shorter summers in the north leads to slower growth and thus later maturation, which could explain why male and female walleyes mature later compared to more southern populations (Scott and Crossman, 1998). Overharvesting of fish results in lower intraspecific competition and which leads to greater food intake per individual and thus faster growth. The faster growth enables fish to achieve maturation at a younger age (Craig *et al*., 1995, Trippel, 1995). Although Waterhen Lake is an exploited fishery (Scott Forbes, pers. comm.), maturation for male and female walleye was found to be similar to those in lakes in and around Manitoba, showing that the fishery has not had an impact on the age-at-maturity of walleye.

This study confirmed that male walleye generally mature one to two years earlier than females (Scott and Crossman, 1998) with 100% of males being mature at age 4 and 100% of females maturing by 6 years (Figure 9).

Once the walleye reached maturity, annual growth increments declined, a trend which was also seen in Babaluk *et al*., 1993, with the exception of the small sample size of the four oldest categories of walleye (five fish aged 7-10 years, Table 6). Decrease in growth rates after maturation has occurred is due to the allocation of energy to reproduce rather than to grow (von Bertalanffy, 1938).

Vulnerability to Mesh Sizes

Data on the vulnerability of different sizes of walleye to different gill-net mesh sizes were gathered by the Manitoba Fisheries Branch over a two-year period (Geoff Klein, pers. comm.) Analysis of this data showed a slightly positive linear relationship between total length and mesh size (Figure 9) but there was a wide range of walleye lengths getting caught in each mesh size.

The relationship between fish size/age and mesh size is complicated by the different modes of fish capture in a gill net. The strongest relationship between mesh size and fish size occurs when fish "gill" in the net – i.e., fish swim headfirst into the net and the mesh encircles the body usually posterior to the gills. However, some fish become entangled around spines or gill covers, and here the relationship between mesh size and fish size is less clear. And in still other cases, very large fish may be captured in very small mesh sizes when they swallow smaller fish trapped in the net.

Thus there are several explanations as to why larger fish could possibly get caught in smaller gill nets. Firstly, they could simply become tangled in the net. As a top predator in freshwater ecosystems, larger walleye could also become caught in smaller mesh sizes by swallowing the net when eating small fish caught in it. These fish could be prey species or even smaller walleyes as it is known to occur that larger walleyes will become cannibalistic (Madenjian *et al*. 1996).

Using the total lengths to estimate age (from mean total length of known-age fish), mesh size recruitment of different ages was produced (Table 4). The trend shows as would be expected: younger fish get caught in smaller nets and older fish get caught in larger nets.

Assessment of Current Harvest Policy

For this fishery to be considered sustainable, the current minimum permitted mesh size of 3.75" (95 mm) must be determined sufficient to prevent harvest of immature walleye in order to avoid recruitment overfishing. Rapid fishery collapses can be brought on by recruitment overfishing when fish are harvested before reaching maturity and thus are unable to spawn before being removed from it. The minimum mesh size of 3.75" (or 95 mm) enforced by the province captured few immature walleye indicating that juveniles were not very susceptible to the fishery (Table 5). Ideally 1–2 years of maturity should pass before fish become vulnerable to the fishery to allow time for a few seasons of reproduction before harvest (Myers and Mertz, 1998). If an evaluation of the fishery finds decreasing numbers of fish reaching maturity in the sample, the harvesting policy will need to be reassessed to allow spawning and reproduction in order to maintain the fish stocks. Age structures thus serve as biological reference points to signal recruitment overfishing (Peterman 2002) and allow fisheries biologists to analyze population dynamics of the fishery. Based on the sample analyzed in this study, the current harvest policy for the walleye fishery at Waterhen Lake, which enforces a minimum mesh size of 3.75", is not harvesting a very large portion of pre-reproductive fish (Table 5). However, in order to ensure maturation and allow spawning at least once from mature fish, this 3.75" mesh may not be sufficient. In order to determine whether or not this 3.75" mesh is a sustainable minimum, a larger sample size of fish lengths and mesh sizes must be analyzed. Keeping a restriction on minimum mesh size will allow this walleye fishery to continue being productive and sustainable without impairing the reproductive capacity of the stock.

Back-calculation of Length-at-Age

Back-calculated lengths-at-age are used to provide additional information on growth of the study species. Lee (1920) described back-calculation as the growth increment of the scale being a constant proportion of the growth increment of the fish. In other words, as the fish length increases so too does the calcified aging structure (be it scales, opercula, spines or vertebrae) will increase in proportion to that. This would allow future workers to collect discarded heads from the commercial fishery to obtain opercula and assess the age and size structure of fish in the harvest.

This "proportional method" of back-calculation method was used in this study and used the length of the fish and the length of the operculum at capture to determine previous lengths-at-age. The back-calculated lengths for walleye from Waterhen Lake were found to consistently underestimate previous lengths-at-age (as determined by mean empirical length at capture data), especially for younger fish (Table 6). Thus this proportional model seems to be viable for back-calculation only for fish over 8 years old where the difference between mean back-calculated lengths and empirical lengths-at-capture are minimal. This result is common in back-calculation, as was found in a study using walleye dorsal spines as back-calculation structures (Borkholder and Edwards, 2001).

Back-calculation would be most accurate if performed during the time of year when differences in growth rates are the lowest which is just prior to annulus formation (Weatherley and Gill, 1987). Annulus formation is likely due to changes in temperature (Babaluk *et al.*, 1993), which is why for walleye in Manitoba annulus formation usually coincides with spawning (but is not directly a result of it as annuli formation occurs in immature walleye as well) in May (Babaluk *et al.*, 1993). Craig (1974) found that perch, which are in the same family as walleye, deposit their annuli with the highest monthly frequency occurring in May. Since fish in this sample were caught in fall, differences found between the back-calculated lengths (lengths at annulus formation) and lengths at capture could be due to somatic growth during the period from the last annulus formation to the time of capture (May-September) This same phenomenon was seen in back-calculated lengths-at-age from Babaluk *et al.* (1993) where fish were collected in the summer and fall seasons.

These back-calculated lengths-at-age must, however, be recognized as simple estimates of age at annulus formation and are not foolproof. Errors could be caused by small things like sampling errors, or too small of a sample size to represent the entire population (Maceina *et al.*, 2007).

This linear relationship is not always the case and is in fact the reason for many errors in back-calculations as not all fish have linear fish length-age structure length ratios over time. Some of these ratios vary with somatic growth rate which leads to large otoliths in slow-growing fish (Campana, 1990) and therefore does not follow a linear relationship. In these cases, using a simple back-calculation as was done in this study is not sufficient, and other calculations would have to be made to account for this (see Campana, 1990 for description of algorithms).

The underestimations of back-calculated lengths-at-age of this data could also be explained by Lee's Phenomenon, which is found in other cases to be the source of error for back-calculation (Klumb *et al*., 1999). This phenomenon is caused by a sampling bias in which the young fish in the sample are represented by only the fast-growing juvenile fish, as the slower-growing fish are too small to be caught in the nets. This causes the empirical size at age to be much larger than that in the wild population if it were to be sampled in its entirety (Carlander, 1997). This low percentage of small sized walleye produces a negative Lee's Phenomenon where the back-calculated lengths-at-age are *smaller* than empirical lengths-at-capture. Table 6 illustrates Lee's Phenomenon when looking down the columns at the back-calculated lengths. The length of a one year old fish is estimated as being much higher from the opercular measurements of a young fish than an old fish. This is well explained by Lee's Phenomenon because that 10-year old fish would have been too small at age one to be caught in the nets (thus why it was able to grow to 10 years) and the one-year old fish is big compared to its age class and thus is being caught in the smaller mesh sizes. Lee's Phenomenon is especially seen in lakes with active fisheries (as opposed to natural, unfished populations) simply due to recruitment. As Ricker (1975) describes, the large fish of a particular year class are more vulnerable than their smaller counterparts, and thus will be far more present in the sample. This is a reasonable explanation as to why there was such a great underestimation of back-calculated lengths-at-age.

Implications for Eco-certification

Since MSC eco-certification requires knowledge of the biological data pertaining to the Waterhen Lake walleye fishery before eco-certification can take place, this study plays an important role in gathering initial assessment of the fishery data. Using the data gathered in this study as a baseline, the five-year assessment period which monitors this biological information (age, size and sex structure of the stock) can now take place. Eco-certification of this small scale freshwater walleye fishery will be novel and important to encourage eco-certification of other freshwater fisheries in Canada. This eco-certification will open the possibility of the fishers marketing the eco-labelled fish at a higher dollar-value and gaining access to this new market which sees suppliers and retailers moving toward a reliance on eco-certified products. A consumer preference for eco-labelled products will keep both the agencies' regulations and industry's compliance with them in check, as the threat of losing certification would cause fisheries to be more precautious by keeping a tighter rein on harvesting regulations (Peterman 2002). The main objective of eco-certification is to reward sustainable fisheries by providing them with a competitive advantage in the marketplace, which is the hope for Waterhen Lake’s walleye fishery.

Conclusions

1) Opercular bones are a valid aging structure for walleye, having a high correlation with ages determined from otoliths, a known valid aging structure.
2) Male walleye from Waterhen Lake mature at an age range of 1-4 years and female walleye mature from 4-6 years and therefore first spawn at age 2-5 years (for males) and 5-7 years (for females).
3) Back-calculation of length-at-age underestimates mean empirical length-at-capture due to Lee's Phenomenon.
4) Current harvest policy at Waterhen Lake which enforces a minimum mesh size of 3.75 inches may be sufficient to prevent recruitment overfishing of the walleye fishery, but analysis of a larger sample size is recommended.
5) With this initial study of the walleye fishery complete, Waterhen Lake could become eco-certified after a five-year assessment period which will determine its sustainability.

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Appendix A. Map of Sample Area – Waterhen Lake, MB.