Cod SD 22 Age reading exchange December 2015

Introduction

In 2014, concerns were raised by WGBFAS when it was noted that the age structure of catches and survey data of eastern Baltic cod were diverging. These concerns echoed the opinions of expert groups who for many years have struggled with standardising the age interpretation methods used for these stocks due to the obscurities observed in the fundamental otolith structure. In 2014, an otolith exchange for Eastern Baltic Cod was initiated by the Thünen Institute of Baltic Sea Fisheries (OF), Rostock, Germany and coordinated and analysed by DTU Aqua National Institute of Aquatic Resources, Denmark. For SD 22, the average percentage agreement was poor at just 70% with a high coefficient of variation of 23.4%. The main issue being that the two German readers estimated the same age while the Danish reader estimated the fish as one year older due to the mis-identification of the first winter ring. However, the result confirmed that the otolith structure observed in cod from SD22 complies with the general requirements for age estimation. Presently, it is the only area within the Baltic from which it is still possible to read cod otoliths by traditional age reading methods. WGBFAS 2015 agreed that a yearly otolith exchange will take place for cod from SD22. The results of the current exchange indicate that the problems with the mis-identification of the first winter ring are almost resolved. The data will be used by WBGFAS 2016 to assess the error in the age information and to implement an age error matrix in the assessment following the general recommendations outlined by WKSABCAL 2014.

The current age calibration exercise was co-ordinated and analysed by Julie Coad Davies of DTU Aqua National Institute of Aquatic Resources, Denmark. Samples were selected by the Thünen Institute of Baltic Sea Fisheries (OF), Rostock, Germany.

Readers

| Reader | Name | Country | Institute and address | e-mail | Experience |
|--------|-----------|---------|----------------------------|-----------------------------|-------------|
| 1 | Svend- | Denmark | DTU Aqua, Charlottenlund | <u>sel@aqua.dtu.dk</u> | Expert |
| | Erik | | Slot, Jægersborg Allé 1, | | |
| | Levinsky | | 2920 Charlottenlund, | | |
| | | | Denmark. | | |
| 2 | Marianna | Germany | Thünen Institute of Baltic | marianna.wolfram@ti.bund.de | Expert |
| | Wolfram | | Sea Fisheries, Alter Hafen | | |
| 3 | Britta | | Süd 2, 18069 Rostock, | britta.preuss@ti.bunde.de | Expert |
| | Preuß | | Germany. | | |
| 4 | Krzysztof | Poland | National Marine Fisheries | kradtke@mir.gdynia.pl | No |
| | Radtke | | Research Institute, | | experience |
| | | | Kołłątaja St. 1, 81-332 | | in cod from |
| | | | Gdynia, Poland. | | SD22 |

Table 1 Participant information

Samples

180 images of sliced otoliths were provided by the Thünen Institute of Baltic Sea Fisheries (OF), Rostock, Germany. One image (21211341_099) was excluded by WebGR during the import of images and otolith data. A range of magnifications were used when taking the images and magnification details were provided for 178 images. The images aimed at covering the widest possible range of ages (age 0 to age 10) and length classes (see below). When selecting the otoliths the priority list from 1-3 was used:

- 1. BITS Q4 2014, 2013, 2012
- 2. Commercial samples from the second half of the year
- 3. Other sources

| Quarter | Month | No. of otoliths |
|---------|----------------|-----------------|
| 4 | October 2011 | 15 |
| 4 | November 2011 | 2 |
| 4 | December 2011 | 1 |
| 4 | October 2012 | 5 |
| 4 | November 2012 | 7 |
| 4 | October 2013 | 13 |
| 4 | December 2013 | 12 |
| 1 | March 2014 | 18 |
| 3 | September 2014 | 3 |
| 4 | October 2014 | 38 |
| 4 | November 2014 | 36 |
| 1 | January 2015 | 30 |

Table 2. Sample information given by number of otoliths per quarter



Figure 1. Distribution of samples within length classes

Methods

Images of sliced otoliths taken under transmitted light using a range of magnifications (x0.5, x0.625, x0.8) were made available for annotation on WebGR. The readers were given an image example showing which axis to annotate and were asked to annotate the nucleus and the start of each winter ring and give a final estimation of age. Readers were provided with information on the capture date, area and total length (TL).

Analysis

The analysis was a 2 step process:

1. Age data

The agreement between readers was analysed by means of the traditional procedures, using the excel workbook "Age Reading Comparisons" (Eltink 2000):

- average % Agreement (nmodal age/ntotal*100)
- coefficient of variation (CV) (Standard deviation/average*100)
- bias plots and tests

Average percentage error was also calculated based on the method outlined by Beamish & Fournier (1981).

2. Measurement of growth zone data

WebGR provides a measure of distance between the annotations made by the readers and thus provides a measure of growth increment width. The "alldistances" dataset from WebGR will be used to compile

growth curves for each fish and for each reader which will be compared by Linear Mixed Effects Models (LMEM's). This analysis will be prepared in time for WGBFAS 2016.

Results

1. Age data

The % agreement (average percentage agreement) between all readers is 91.9% with a CV (coefficient of variation) of 9.9%. When only the 3 expert readers (from Denmark and Germany) are included, the % agreement increases to 94.4% and the CV decreases to 2.8%.

As Reader 4 (from Poland) does not routinely read cod from SD22 the following results are based on only the 3 expert readers (DK, GER_M and GER_B). The results including all readers are shown in Tables 3a – 7a and Figure 2a and can be found at the end of the report.

Tables 3-7 show the age composition, % agreement by modal age, CV by modal age, Relative bias by modal age and inter reader bias tests by modal age respectively. Figure 2 shows the age bias plots for each reader and all expert readers combined. For ages 0 and 1 there is 100% agreement (Table 4) between the expert readers and it appears that all readers are annotating the same structures and thus there are no problems with the identification of the first winter ring in the youngest fish. From age 2-4 the % agreement decreases to between 94.7 - 97.3 % and the CV (Table 5) increases from 6.9 - 1.8%. This is mostly due to Reader DK not counting the first winter; see Figure 2 where Reader DK estimates an age of 2 and Readers GER_M and GER_B estimates an age of 3, and possibly due to GER_M omitting to enter an age in WebGR. From age 6 upwards the % agreement is generally lower (66.7 - 95%) and the CV generally higher (6.4 - 1.3%). Again this is mostly due to difficulties in identifying the first winter ring; see Figure 3 where Reader DK and GER_B estimate an age of 6 and Reader GER_M estimates and age of 7.

The average percentage error (APE) can better quantify the precision in the age readings as it takes the age of the fish into consideration when estimating the error in the age readings. The overall APE of 1.6% (expert readers only) indicates a very high level of precision between readers.

| Age | DK | GER_M | GER_B | Total |
|-------|-----|-------|-------|-------|
| 0 | 25 | 26 | 25 | 76 |
| 1 | 25 | 24 | 25 | 74 |
| 2 | 29 | 23 | 25 | 77 |
| 3 | 23 | 25 | 25 | 73 |
| 4 | 27 | 25 | 24 | 76 |
| 5 | 15 | 14 | 14 | 43 |
| 6 | 11 | 12 | 12 | 35 |
| 7 | 17 | 21 | 22 | 60 |
| 8 | 3 | 2 | 1 | 6 |
| 9 | 1 | 2 | 2 | 5 |
| 10 | 1 | 2 | 1 | 4 |
| 11 | 2 | 1 | 3 | 6 |
| Total | 179 | 177 | 179 | 535 |

Table 3. Age composition (based on estimated ages) and the actual number of otoliths read



Figure 2. Image 21411431_094. Reader DK, age 2 (bright green); Reader GER_M, age 3 (blue) and Reader GER_B, age 3 (dark green)



Figure 3. Image 21403310_018. Reader DK, age 6 (bright green); Reader GER_M, age 6 (blue) and Reader GER_B, age 7 (dark green)

| MODAL age | DK | GER_M | GER_B | Expert Readers |
|---------------|-------|-------|-------|-------------------|
| 0 | 100% | 100% | 100% | 100.0% |
| 1 | 100% | 100% | 100% | 100.0% |
| 2 | 100% | 92% | 100% | 97.3% |
| 3 | 84% | 100% | 100% | 94.7% |
| 4 | 92% | 100% | 96% | 96.0% |
| 5 | 71% | 100% | 93% | 88.1% |
| 6 | 62% | 92% | 85% | 79.5% |
| 7 | 85% | 100% | 100% | 95.0% |
| 8 | 100% | 100% | 50% | 83.3% |
| 9 | 50% | 100% | 50% | 66.7% |
| 10 | 100% | 100% | 0% | 66.7% |
| 11 | 100% | 50% | 100% | 83.3% |
| Weighted mean | 89.4% | 97.7% | 96.1% | 01 10/ |
| Ranking | 3 | 1 | 2 | 34.4% |

Table 4. Percentage agreement for each reader by modal age

| MODAL age | DK | GER_M | GER_B | Expert Readers |
|---------------|------|-------|-------|-------------------|
| 0 | 0% | 0% | 0% | 0.0% |
| 1 | 0% | 0% | 0% | 0.0% |
| 2 | 0% | 30% | 0% | 6.9% |
| 3 | 13% | 0% | 0% | 3.5% |
| 4 | 7% | 0% | 5% | 1.8% |
| 5 | 10% | 0% | 5% | 4.3% |
| 6 | 9% | 5% | 6% | 6.0% |
| 7 | 5% | 0% | 0% | 1.3% |
| 8 | 0% | 0% | 8% | 3.5% |
| 9 | 8% | 0% | 7% | 6.4% |
| 10 | - | - | - | - |
| 11 | 0% | 7% | 0% | 2.7% |
| Weighted mean | 4.9% | 4.7% | 1.7% | 2 00/ |
| Ranking | 3 | 2 | 1 | 2.0% |

Table 5. Co-efficient of Variation (CV) for each reader by modal age

The overall relative bias value is -0.03 which shows that the readers are generally underestimating the age of the fish in comparison to the modal age. This is mostly due to the tendency for Reader DK to omit the first winter ring. Reader GER_B is more likely to overestimate the ages. The age bias plots in Figure 4 show the results from the calculations of relative bias by modal age (Table 6) clearly. Table 7 shows the inter reader bias test and reader against modal age bias test and it is only between GER_M and modal age that there is no possibility of bias.

| MODAL age | DK | GER_M | GER_B | Expert Readers |
|---------------|-------|-------|-------|-------------------|
| 0 | 0.00 | 0.00 | 0.00 | 0.00 |
| 1 | 0.00 | 0.00 | 0.00 | 0.00 |
| 2 | 0.00 | -0.16 | 0.00 | -0.05 |
| 3 | -0.16 | 0.00 | 0.00 | -0.05 |
| 4 | -0.08 | 0.00 | 0.04 | -0.01 |
| 5 | -0.29 | 0.00 | 0.07 | -0.07 |
| 6 | -0.38 | 0.08 | 0.15 | -0.05 |
| 7 | -0.15 | 0.00 | 0.00 | -0.05 |
| 8 | 0.00 | 0.00 | 0.50 | 0.17 |
| 9 | -0.50 | 0.00 | 0.50 | 0.00 |
| 10 | 0.00 | 0.00 | 1.00 | 0.33 |
| 11 | 0.00 | -0.50 | 0.00 | -0.17 |
| Weighted mean | -0.11 | -0.02 | 0.04 | 0.02 |
| Ranking | 3 | 1 | 2 | -0.03 |

Table 6. Relative Bias for each reader by modal age

| | DK | GER_M | GER_B |
|-----------|-----|-------|-------|
| DK | | * | * * |
| GER_M | * | | * |
| GER_B | * * | * | |
| Modal age | * * | | * |

Table 7. Inter reader bias test and reader against modal age bias test





Figure 4. Age bias plots as mean age \pm 2 SD on modal age for each expert reader and all experts combined

Problems with identification of the edge were only apparent with the inexperienced reader from Poland who in some examples of 0 year old fish counted an extra ring at the edge (this was not a consistent problem).

2. Measurement of growth zone data

Once the input error in WebGR has been resolved it should be possible to standardize the measurements between annotations made on images taken using the various magnifications.

Conclusions

These results show an improvement on the 2014 exercise where the % agreement between the same 3 readers was 70% and is now 94.4% and where the CV was 23.4% and is now 2.8%. In the 2014 exchange the main problem identified was that the two German readers were estimating the same age while the Danish reader was estimating the fish as one year older due to misinterpretation of the juvenile settling ring as a true winter ring. This issue now appears to be almost resolved. For ages 0 and 1 there is 100% agreement between the expert readers. From age 2 and upwards there are some otoliths where the growth structures in the centre of the otoliths are not so well defined and this leads to the disagreement as to where the first true winter ring is found.

The inexperienced reader does not provide age estimates of cod from SD 22 for assessment purposes. However, when their readings are included in the analysis the overall performance of the group is lowered. Thus this reader should consider the results of this exchange when reading cod otoliths from other areas. The analysis of the measurement data should be able to a) confirm whether the readers are in agreement as to which structures are the winter rings and b) provide a range of distances from the centre to the first winter ring which can be used as a guideline for the readers.

Results with Reader 4 (POL) included

When looking at the age estimates of Reader 4; some of the modal age 0 otoliths were estimated to be 0 years old while some were estimated to be 1 year old. Given that these otoliths are almost identical in appearance and caught at the same time of year these age estimates should be interpreted with caution. In general, Reader 4's ages are overestimated in comparison with the modal age and the other readers, see Table 6a and the corresponding Figure 2a.

| Age | DK | GER_M | GER_B | POL | Total |
|-------|-----|-------|-------|-----|-------|
| 0 | 25 | 26 | 25 | 19 | 95 |
| 1 | 25 | 24 | 25 | 31 | 105 |
| 2 | 29 | 23 | 25 | 23 | 100 |
| 3 | 23 | 25 | 25 | 26 | 99 |
| 4 | 27 | 25 | 24 | 18 | 94 |
| 5 | 15 | 14 | 14 | 20 | 63 |
| 6 | 11 | 12 | 12 | 12 | 47 |
| 7 | 17 | 21 | 22 | 17 | 77 |
| 8 | 3 | 2 | 1 | 5 | 11 |
| 9 | 1 | 2 | 2 | 3 | 8 |
| 10 | 1 | 2 | 1 | 4 | 8 |
| 11 | 2 | 1 | 3 | - | 6 |
| Total | 179 | 177 | 179 | 178 | 713 |

Table 3a. Age composition (based on estimated ages) and the number of otoliths read

Table 4a. Percentage agreement for each of the readers by modal age

| MODAL age | DK | GER_M | GER_B | POL | All |
|---------------|-------|-------|-------|--------|---------|
| | | | | | Readers |
| 0 | 100% | 100% | 100% | 76% | 93.9% |
| 1 | 100% | 100% | 100% | 100% | 100.0% |
| 2 | 100% | 92% | 100% | 96% | 97.0% |
| 3 | 84% | 100% | 100% | 100% | 96.0% |
| 4 | 92% | 100% | 96% | 72% | 90.0% |
| 5 | 71% | 100% | 93% | 93% | 89.3% |
| 6 | 62% | 92% | 85% | 85% | 80.8% |
| 7 | 85% | 100% | 100% | 70% | 88.8% |
| 8 | 100% | 100% | 50% | 0% | 62.5% |
| 9 | 50% | 100% | 50% | 50% | 62.5% |
| 10 | 100% | 100% | 0% | 100% | 75.0% |
| 11 | 100% | 50% | 100% | 0% | 62.5% |
| Weighted mean | 89.4% | 97.7% | 96.1% | 84.30% | 01 00/ |
| Ranking | 3 | 1 | 2 | 4 | 91.9% |

| MODAL age | DK | GER_M | GER_B | POL | All Readers |
|---------------|------|-------|-------|-------|----------------|
| 0 | 0% | 0% | 0% | 182% | 48.0% |
| 1 | 0% | 0% | 0% | 0% | 0.0% |
| 2 | 0% | 30% | 0% | 10% | 6.2% |
| 3 | 13% | 0% | 0% | 0% | 2.9% |
| 4 | 7% | 0% | 5% | 11% | 4.4% |
| 5 | 10% | 0% | 5% | 5% | 4.4% |
| 6 | 9% | 5% | 6% | 6% | 5.3% |
| 7 | 5% | 0% | 0% | 8% | 3.1% |
| 8 | 0% | 0% | 8% | 18% | 6.6% |
| 9 | 8% | 0% | 7% | 7% | 5.9% |
| 10 | - | - | - | - | - |
| 11 | 0% | 7% | 0% | 0% | 5.1% |
| Weighted mean | 4.9% | 4.7% | 1.7% | 30.4% | 0.0% |
| Ranking | 3 | 2 | 1 | 4 | 9.9% |

Table 5a. Co-efficient of Variation (CV) for each of the readers by modal age

Table 6a. Relative Bias for each of the readers by modal age

| MODAL age | DK | GER_M | GER_B | POL | All Readers | |
|---------------|-------|-------|-------|-------|----------------|--|
| 0 | 0.00 | 0.00 | 0.00 | 0.24 | 0.06 | |
| 1 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | |
| 2 | 0.00 | -0.16 | 0.00 | 0.04 | -0.03 | |
| 3 | -0.16 | 0.00 | 0.00 | 0.00 | -0.04 | |
| 4 | -0.08 | 0.00 | 0.04 | 0.28 | 0.06 | |
| 5 | -0.29 | 0.00 | 0.07 | 0.07 | -0.04 | |
| 6 | -0.38 | 0.08 | 0.15 | 0.15 | 0.00 | |
| 7 | -0.15 | 0.00 | 0.00 | 0.35 | 0.05 | |
| 8 | 0.00 | 0.00 | 0.50 | 0.00 | 0.13 | |
| 9 | -0.50 | 0.00 | 0.50 | 0.50 | 0.13 | |
| 10 | 0.00 | 0.00 | 1.00 | 0.00 | 0.25 | |
| 11 | 0.00 | -0.50 | 0.00 | -1.00 | -0.38 | |
| Weighted mean | -0.11 | -0.02 | 0.04 | 0.13 | 0.01 | |
| Ranking | 3 | 1 | 2 | 4 | 0.01 | |

| | DK | GER_M | GER_B | POL | = no sign of bias (p>0.05) |
|-----------|-----|-------|-------|-----|--|
| DK | | * | * * | * * | # = possibility of bias (0.01<p<0.05)< li=""></p<0.05)<> |
| GER_M | * | | * | * * | <pre>* * = certainty of bias (p<0.01)</pre> |
| GER_B | * * | * | | * * | |
| POL | * * | * * | * * | | |
| Modal age | * * | | * | * * | |

Table 7a. Inter reader bias test and reader against modal age bias test



Figure 2a. Age bias plot as mean age ± 2 SD on modal age for Reader 4

References

Beamish, R. J. and Fournier, D. A. (1981) A method for comparing the precision of a set of age determinations. Can. J. Fish. Aquat. Sci. **38**: pp: 982-983

Eltink, A.T.G.W. 2000. Age reading comparisons. (MS Excel workbook version 1.0 October 2000).

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