

# **Stichting DLO Centre for Fisheries Research (CVO)**

## **Report of the Horse Mackerel Exchange and Workshop 2006**

Loes J. Bolle, Pablo Abaunza, Cornelia Albrecht, André Dijkman-Dulkes, Clara Dueñas, Gudrun Gentschouw, Helga Gill, Gert Holst, Ana Moreira, Eugene Mullins, Iñaki Rico, Simon Rijs, Turloch Smith, Aage Thaarup, Jens Ulleweit

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Stichting DLO Centre for Fisheries Research (CVO) P.O. Box 68 1970 AB IJmuiden Phone. +31 (0)317-487418 Fax. +31 (0)317-487326

Visitor address: Haringkade 1 1976 CP IJmuiden

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#### Summary

Following a recommendation from PGCCDBS, a workshop on age calibration of horse mackerel was carried out. The workshop was preceded by an exchange. The objectives were:

- a. In general, improve the quality of horse mackerel readings by international calibration.
- b. In particular, attempt to resolve the observed differences between countries.
- c. Estimate the accuracy and precision of the age readings before and after the intercalibration.
- d. Take into account differences between areas and methods.
- e. Training of new horse mackerel readers.

Eight experienced readers participated in the exchange, 7 of which also participated in the workshop. Five trainees participated in the workshop, only one of them also participated in the exchange. All countries providing age reading data to the WGMHSA were represented in both the exchange and the workshop by an experienced reader.

Portugal, Germany and The Netherlands provided otolith sets for the exchange. The 8 sets represented different otolith preparation methods and stocks. Three sets consisted of otoliths from the extremely strong 1982 year-class and hence the age is considered to be known. One set focused on the young fish, as these were expected to present problems based on an informal small-scale otolith exchange.

Horse mackerel is regarded to be a difficult species to age and this is reflected by the results of the exchange. The agreement between the experienced readers was low, especially for otoliths from the Southern stock. For the 'known-age' sets, agreement with the modal age was higher than with 'true' age. Comparison with the 'true' ages showed an overall tendency to underestimate age. The experienced readers were accustomed to different otolith preparation methods and different growth patterns associated with the different stocks. Generally, the readers had more difficulty if they were reading material they were not accustomed to.

For 3 sets containing Western stock otoliths, digital images of the sectioned otoliths were annotated by the readers participating in the exchange. During the workshop these annotated images were used to discuss differences in interpretation. A great deal of attention was paid to the interpretation of the first annuli, both in young fish as well as in older fish. This point appeared to be the major cause of differences in interpretation. In some otoliths split rings or the interpretation of the edge of the otoliths caused problems. All these features were discussed and eventually consensus was reached for all otoliths put up on the screen. For a small subset of the Southern stock otoliths, images of sectioned otoliths were digitised during the meeting. These images were discussed in the group. In some cases consensus could be reached on how to interpret the otolith, however in other cases it seemed to be impossible to age the otolith. Ageing of the Southern stock otoliths appeared to be less difficult when using broken-burnt material instead of (images of) sectioned otoliths.

Two new otoliths sets were presented to the readers during the workshop. These sets were designed to be identical (in size range, age composition and catch months) to 2 sets included in the exchange. At the end of the workshop all readers re-read these 2 exchange sets. The results of the 7 experienced readers clearly showed an improvement from exchange sets to the workshop sets, and from the workshop sets to the reread of the exchange sets. Although it can be argued that the readers may have remembered their first age readings, this seems unlikely because the second reading was carried out 2–7 months later and the readers were not informed that they were re-reading exchange sets. Most of the trainees only participated in the workshop, so the workshop sets were the first sets for them to read. Comparison of their results for workshop sets and the consecutive reading of the exchanges sets showed a tremendous improvement.

#### 1. Introduction

#### 1.1 Background

The ICES Planning Group on Commercial Catch, Discards and Biological Sampling (PGCCDBS) meeting in 2004 (ICES, 2004) identified horse mackerel as one of the species which required international age reading calibration. The last workshop had been held in 1999 (ICES, 1999). Furthermore, indications for age reading problems existed. At the 2004 meeting of the Working Group on the Assessment of Mackerel, Horse Mackerel Sardine and Anchovy (WGMHSA) differences between the age compositions of Dutch and German samples collected in Divisions VIId,e,h were observed (ICES, 2005). Furthermore, an informal small-scale otolith exchange indicated age reading differences between 4 readers from 4 different countries.

The horse mackerel exchange was originally planned for 2005, but due to logistic problems it was postponed until 2006. The workshop was held from 12 to 14 December 2006 in IJmuiden, The Netherlands.

#### **1.2** Horse mackerel stocks

The Working Group on the Assessment of Mackerel, Horse Mackerel Sardine and Anchovy (WGMHSA) distinguishes 3 stocks in the north-east Atlantic: the North Sea stock, the Western stock and the Southern stock (Figure 1). The catches and sampling levels by country in 2004 are presented in Table 1.

(1013, 2003).	Official		Number of	Number	Number
	catch (t)	sampling (%)	samples	measured	aged
Netherlands	67,289	93	80	11,615	2,000
Spain	28,147	98	527	43,097	3,413
Ireland	26,432	77	31	5,121	1,827
Germany	22,742	59	57	17,953	2,255
Denmark	20,267	0	0	0	0
Portugal	11,875	100	964	133,534	1,582
Norw ay	10,751	98	13	1,746	393
France	10,590	0	0	0	0
UK (Eng. & Wal.)	10,251	0	0	0	0
Faroe	3,849	0	0	0	0
UK (Scotland)	1,524	0	0	0	0
Sw eden	665	0	0	0	0
Russia	5	0	0	0	0
Belgium	4	0	0	0	0
Total	216,361	68	1,672	213,066	11,470

Table 1. Catches and sampling levels by country for all horse mackerel stocks combined in 2004 (ICES, 2005).

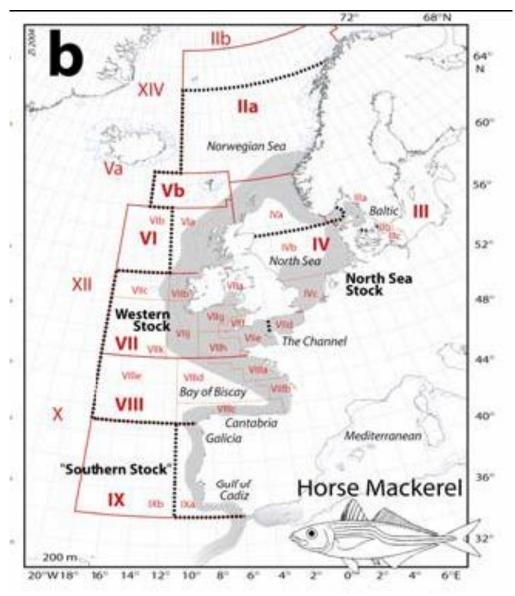


Figure 1. The horse mackerel stocks assessed within the Working Group on the Assessment of Mackerel, Horse Mackerel Sardine and Anchovy (ICES, 2005).

#### 1.3 Objectives

The objectives of the exchange and workshop were:

- a. In general, improve the quality of horse mackerel readings by international calibration.
- b. In particular, attempt to resolve the observed differences between countries.
- c. Estimate the accuracy and precision of the age readings before and after the intercalibration.
- d. Take into account differences between areas and methods.
- e. Training of new horse mackerel readers.

#### 2. Methods

#### 2.1 Participants

The participants in exchange and workshop are listed in Table 2. Eight experienced age readers participated in the exchange, 7 of which also participated in the workshop. Five trainees participated in the workshop, only one of them also participated in the exchange. Three age coordinators attended the workshop. All countries providing age reading data to WGMHSA were represented in both the exchange and the workshop by an experienced age reader (Tables 1 and 2).

Country (institute)	Participant	Email address	Exchange	Workshop	Expertise
Netherlands (IMARES)	Simon Rijs	simon.rijs@w ur.nl	yes	yes	experienced
	André Dijkman	andre.dijkman@w ur.nl	yes	yes	trainee
	Loes Bolle	loes.bolle@wur.nl	no	yes	-
Portugal (IPIMAR)	Ana Moreira	amoreira@ipimar.pt	yes	yes	experienced
	Maria João Ferreira	mjferr@ipimar.pt	yes	no	experienced
Germany (BFA)	Gudrun Gentschow	gudrun.gentschow @ish.bfa-fish.de	yes	yes	experienced
	Cornelia Albrecht	cornelia.albrecht@ish.bfa-fisch.de	no	yes	trainee
	Jens Ullew eit	jens.ullew eit@ish.bfa-fisch.de	no	yes	-
Denemarken (DIFRES)	Aage Thaarup	att@difres.dk	yes	yes	experienced
	Gert Holst	gho@difres.dk	no	yes	trainee
Norway (IMR)	Helga Gill	helga@imr.no	yes	yes	experienced
Spain (AZTI)	lñaki Rico	irico@pas.azti.es	yes	yes	experienced
Spain (IEO)	Clara Dueñas	clara@st.ieo.es	no	yes	trainee
	Pablo Abaunza	pablo.abaunza@st.ieo.es	no	yes	-
Ireland (Marine Institute)	Eugene Mullins	eugene.mullins@marine.ie	yes	yes	experienced
	Turloch Smith	turloch.smith@marine.ie	no	yes	trainee

Table 2. Participants in the exchange and workshop.

Most of the participating countries age fish from the Western stock. Some also age North Sea stock fish, besides Western stock fish. Only Portugal and one of the 2 Spanish institutes age fish from the Southern stock (Table 3). Horse mackerel from the Southern stock is considered to be more difficult to age than horse mackerel from the other 2 stocks.

#### 2.2 Age reading methods

Different preparation methods are used by the different laboratories (Table 3). Most laboratories age horse mackerel using broken-burnt otoliths, despite the fact that transverse sections was recommended by the previous workshop (ICES, 1999).

Country (institute)	Stocks	Preparation method
Netherlands (IMARES)	Western & North Sea stocks	sections
Portugal (IPIMAR)	Southern stock	w hole for smaller fish, broken-burnt for larger fish
Germany (BFA)	Western & North Sea stocks	stained sections
Denemarken (DIFRES)	Western & North Sea stocks	w hole for smaller fish, broken-burnt for larger fish
Norw ay (IMR)	Western stock	broken-burnt
Spain (AZTI)	Western stock	broken-burnt
Spain (IEO)	Southern stock	broken-burnt
Ireland (Marine Institute)	Western stock	stained sections

Table 3 Stocks sampled and	preparation methods used b	v the different laboratories
Table 5. Stocks sampled and	preparation methods used b	y the unrelent laboratories.

Netherlands: One otolith of the pair is sectioned. Otoliths are embedded in black resin. Transverse sections ( $\pm 0.5$  mm) are mounted onto glass slides. Sections used to be stained with 'Honey Pine' wood finisher, but this procedure was abandoned as it appears to cause deterioration of the material on the long term. The otoliths are read using reflected light. The translucent bands are counted.

Germany: Same procedures as the Netherlands, except the sections are stained ('Honey Pine' wood finisher) and mounted onto glass slides after ageing.

Ireland: Same procedures as the Netherlands, except the sections are stained ('Honey Pine' wood finisher).

Portugal: Smaller fish (<35 cm) are aged using whole otoliths. The otoliths are soaked in a tymol 5% solution for 1 day after which the otoliths are aged using emersion oil and reflected light. Larger fish ( $\geq$ 35 cm) are aged using broken-burnt otoliths. The translucent bands are counted.

Denmark: Same procedure as Portugal, except the whole otoliths are submerged in water or alcohol.

Spain: One otolith of the pair is broken through the nucleus. The broken otolith is burnt using a thermostat regulated hotplate. The burnt half otolith is mounted in black plasticine and submerged in 70% alcohol, together with the untreated whole otolith. Both are read using reflected light. The translucent bands are counted.

Norway: One otolith of the pair is broken through the nucleus. The broken otolith is burnt. The otolith is read using baby oil on the surface and reflected light. The translucent bands are counted.

#### 2.3 Otoliths sets

Portugal, Germany and The Netherlands provided otolith sets for the exchange. The sets represented different otolith preparation methods and stocks (Table 4). Sets G & K consisted of otoliths from the extremely strong 1982 year-class and hence the age is considered to be known (with a certainty of approximately 95%). These sets have been used in the previous horse mackerel workshop (ICES, 1999) and were therefore renumbered for this exchange. Set NL-VIIe-2003 focused on young fish, as these were expected to present problems based on the findings of an informal small-scale otolith exchange.

For all otoliths sets, except the Portuguese set, a subset of the complete otolith set supplied was used in the exchange. For the Portuguese set, one otolith of the pair was sectioned, the other otolith was either whole (fish length <35 cm) or broken burnt (fish length  $\geq$ 35 cm). For the Dutch set G, one otolith of the pair was sectioned, the other was broken-burnt. All other sets consisted of sectioned otoliths. A total of 357 otoliths were included in the exchange (Table 4).

A second subset was extracted from set G and presented to the readers during the workshop, to examine if the discussions on the interpretation of otoliths had led to a higher agreement between the readers. Both the exchange subset (G1) and the workshop subset (G2) consisted of 4–5 fish per age group in the age range of 4 to 13 ('true' ages). Both subsets were also comparable in size distribution (24–35 cm) and catch months (mainly January-March, some from April-May). Likewise, a second subset was extracted from the Dutch set containing only young fish (NL-VIIe-2003). Only sectioned otoliths were included in these workshop sets (Table 4).

Country	Otolith set	Area	Stock	Months	Size range	Preparation method	N (total)	N (exc)	N (w k)
Portugal	PT-IXa-2005	ICES IXa	Southern	1-3	21-42 cm	section	51	50	
						w hole or broken-burnt	51	51	
Germany	DE-IV-2005	ICES IV	North Sea	7-8	19-33 cm	stained section	175	48	
Germany	DE-V11d-2005	ICES V IId	Western	10	18-37 cm	stained section	233	51	
Netherlands	NL-V1le-2003	ICES V lle	Western	8	16-20 cm	section	100	23	10
Netherlands	G	ICES V II	Western	1-5	24-35 cm	section	170	48	48
						broken-burnt	170	48	
Netherlands	К	ICES V II	Western	9-11	14-30 cm	stained section	153	38	

Table 4. Otolith sets included in exchange and workshop

#### 2.4 Age readings

The exchange sets were aged prior to the workshop. Not all material was aged by all readers: reader 9 did not age the broken-burnt otoliths, readers 2 and 8 did not age the sections of the Portuguese otolith set, and several readers did not age all otoliths in the Portuguese set (Tables 6-8).

Images of sectioned otoliths were available for the 3 Dutch sets (set G, set K and set NL-VIIe-2003) during the exchange. All exchange participants annotated these images prior to the workshop. For a small selection of the Portuguese otoliths, images of sectioned otoliths were made during the workshop. These (annotated) images were used in the workshop to discuss differences in interpretation. Several features (such as first annulus, split rings, otolith edge), which may cause differences in interpretation were discussed during the workshop.

After the first round of discussions, new otolith subsets (see Table 4 and section 2.3) were presented to the readers. After the second round of discussions the readers re-aged otoliths included in the exchange: set G (both broken-burnt as well as sectioned otoliths) and part of set NL-VIIe-2003.

#### 2.5 Data analysis

Data analysis was done, for each set and preparation method separately, using the Age Reading Comparisons tool in Excel (Eltink et al. 2000).

Individual age readings were either compared to modal age or 'true' age. For otolith sets G and K, the age is considered to be known, because these sets consisted of otoliths from the extremely strong 1982 year-class. True age was unknown for the other sets, therefore the age readings were compared to modal age. The modal age estimate was based on the age readings of the 7 experienced age readers who participated in both the exchange and the workshop (readers 1-7 in the tables).

#### 3. Results

#### 3.1 Exchange results

Horse mackerel is regarded to be a difficult species to age and this is reflected by the results of the exchange (Tables 5-8). Overall agreement with modal age, based on experienced readers only, was low for otoliths from the North Sea and Western stocks (53-68%, Table 5), and very low for otoliths from the Southern stock (37-38%, Table 5).

For sets G and K, the agreement with the modal age was higher than with 'true' age (Table 5). Comparison with the 'true' ages showed an overall tendency to underestimate age (top left panel in Figure 5).

Differences between readers and between otolith sets were evident (Tables 6-8). Furthermore, a good reader for one otolith set may perform badly for another otolith set. For example, reader 2 achieved a relatively high agreement (71%) with modal age for the set DE-IV-2005, but a low agreement (16%) for set DE-VIId-2005, because of an overestimation by approximately 1 year (Table 6). Another example is reader 4, who achieved relatively high agreements in both German sets (65-71%), but a low agreement (13%) for set NL-VIIe-2003, because of an underestimation by approximately 1 year (Table 6). These differences by set provided indications where the differences in interpretation may lie. In the first example, the interpretation of the edge in relation to the catch month may differ between reader 2 and the other readers. In the second example, the first annulus is most probably the cause of interpretation differences.

Overall, the results for sectioned otoliths do not appear to be better or worse than the results for brokenburnt otoliths (Table 5). However, the preparation method can make a great difference for the individual reader. For example, see the results of reader 1 and 5 for set G (Table 7). Reader 1 had 67% agreement for sections and 42% for broken-burnt, whereas reader 5 had 48% agreement for sections and 67% for broken-burnt otoliths.

The experienced readers were accustomed to different otolith preparation methods and different growth patterns associated with the different stocks. Generally, the readers had more difficulty if they were reading material they were not accustomed to.

Otolith set (preparation method)	5 5	eement	
Cloint set (preparation method)	IN	'true' age	modal age
PT-IXa-2005 (w hole/broken-burnt)	51	n.a.	38%
PT-IXa-2005 (sections)	50	n.a.	37%
DE-IV-2005 (sections)	48	n.a.	62%
DE-VIId-2005 (sections)	51	n.a.	58%
NL-VIIe-2003 (sections)	23	n.a.	68%
G (broken-burnt)	48	39%	54%
G (sections)	48	43%	53%
K (sections)	38	36%	56%

Table 5. Overall agreement with modal age and 'true' age for the 7 experienced readers who participated in both the workshop and the exchange.

Otolith set (preparation method)		Reader 1	Reader 2	Reader 3	Reader 4	Reader 5	Reader 6	Reader 7	Reader 8	Reader 9
DE-IV-2005 (sections)	N aged	48	48	48	48	48	48	48	48	48
	Agreement	71%	71%	69%	65%	40%	67%	54%	35%	8%
	CV	10%	10%	12%	10%	15%	14%	13%	20%	12%
	Bias	0.2	0.2	0.3	-0.3	-0.6	0.0	-0.3	0.5	-1.1
	Rank	1	2	5	3	8	4	6	9	7
DE-VIId-2005 (sections)	N aged	51	51	51	51	51	51	51	51	51
	Agreement	75%	16%	49%	71%	63%	71%	39%	27%	41%
	CV	6%	10%	9%	7%	10%	9%	13%	21%	13%
	Bias	0.0	0.9	0.4	0.0	-0.3	-0.2	-0.2	0.6	-0.3
	Rank	1	8	5	2	4	3	6	9	7
NL-VIIe-2003 (sections)	N aged	23	23	23	23	23	23	23	23	23
	Agreement	100%	91%	96%	13%	65%	96%	13%	57%	83%
	CV	0%	10%	0%	26%	29%	0%	14%	34%	18%
	Bias	0.0	0.1	0.0	-0.9	-0.3	0.0	1.0	-0.3	-0.1
	Rank	1	4	2	9	6	2	7	7	5

#### Table 6. Western and North Sea stock – Comparison with modal age of 7 experienced readers.

#### Table 7. Western stock – Comparison with 'true' age.

Otolith set (preparation method)		Reader 1	Reader 2	Reader 3	Reader 4	Reader 5	Reader 6	Reader 7	Reader 8	Reader 9
G (sections)	N aged	48	48	48	48	48	48	48	48	48
	Agreement	67%	60%	25%	56%	48%	38%	8%	29%	44%
	CV	7%	10%	11%	11%	7%	10%	13%	12%	8%
	Bias	0.2	0.0	-1.3	0.3	-0.6	-0.9	-1.8	0.6	-0.4
	Rank	1	2	8	4	3	6	9	7	4
G (broken-burnt)	N aged	48	48	48	48	48	48	48	48	0
	Agreement	42%	44%	25%	33%	67%	31%	31%	27%	-
	CV	10%	7%	10%	18%	6%	12%	18%	13%	-
	Bias	-0.7	-0.6	-1.8	0.2	-0.3	-1.3	-1.2	-0.5	-
	Rank	3	2	8	4	1	6	7	5	-
K (sections)	N aged	38	38	38	38	38	38	38	38	38
	Agreement	37%	47%	16%	24%	58%	39%	29%	32%	29%
	CV	5%	13%	16%	14%	9%	12%	16%	28%	10%
	Bias	-0.8	-0.4	-1.9	-1.3	-0.4	-0.9	-1.4	-1.4	-1.1
	Rank	2	3	9	6	1	4	6	8	5

#### Table 8. Southern stock – Comparison with modal age of 7 experienced readers.

Otolith set (preparation method)		Reader 1	Reader 2	Reader 3	Reader 4	Reader 5	Reader 6	Reader 7	Reader 8	Reader 9
PT-IXa-2005 (sections)	N aged	24	0	50	50	50	50	50	0	50
	Agreement	50%	-	20%	20%	54%	38%	42%	-	10%
	CV	9%	-	12%	13%	14%	12%	10%	-	22%
	Bias	0.7	-	-1.3	1.7	-0.1	-0.6	0.5	-	5.3
	Rank	1	-	5	6	3	4	1	-	7
PT-IXa-2005 (w hole/broken-burnt)	N aged	32	30	49	33	29	49	51	51	0
	Agreement	53%	37%	29%	27%	31%	45%	37%	4%	-
	CV	7%	9%	11%	14%	11%	9%	10%	20%	-
	Bias	0.5	1.6	-1.4	2.3	0.4	-1.0	0.9	3.3	-
	Rank	1	5	6	7	4	2	3	8	-

#### 3.2 Workshop discussions

For the Western stock, images were annotated during the exchange and these were used in the workshop to discuss differences in interpretation. A great deal of attention was paid to the interpretation of the first annuli, both in young fish as well as in older fish (Figure 2-3). This point appeared to be the major cause of differences in interpretation. In some otoliths split rings (Figure 4) or the interpretation of the edge of the otoliths caused problems. All these features were discussed and eventually consensus was reached for all otoliths put up on the screen.

For the Southern stock, images of sectioned otoliths were digitised during the meeting. These images were discussed in the group. In some cases consensus could be reached on how to interpret the otolith, however in other cases it seemed to be impossible to age the otolith. Broken-burnt material was also reexamined during the group discussions. Ageing of the Southern stock otoliths appeared to be less difficult when using broken-burnt material instead of (images of) sectioned otoliths. However, this was not an evident result in the exchange (Table 5).

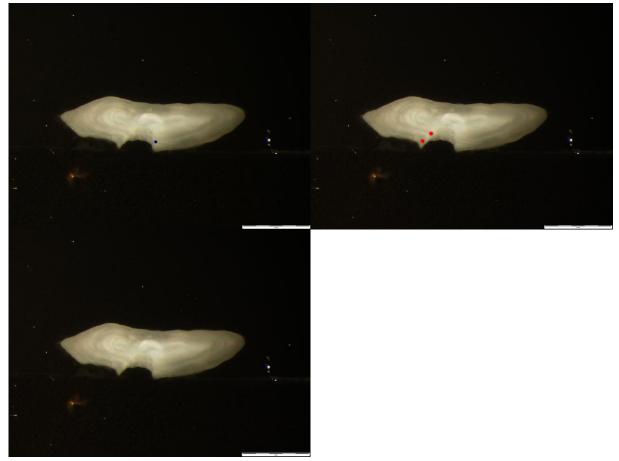


Figure 2. Three different interpretations of an otolith (18.6 cm, August, VIIe). The agreed interpretation is 2 years old.

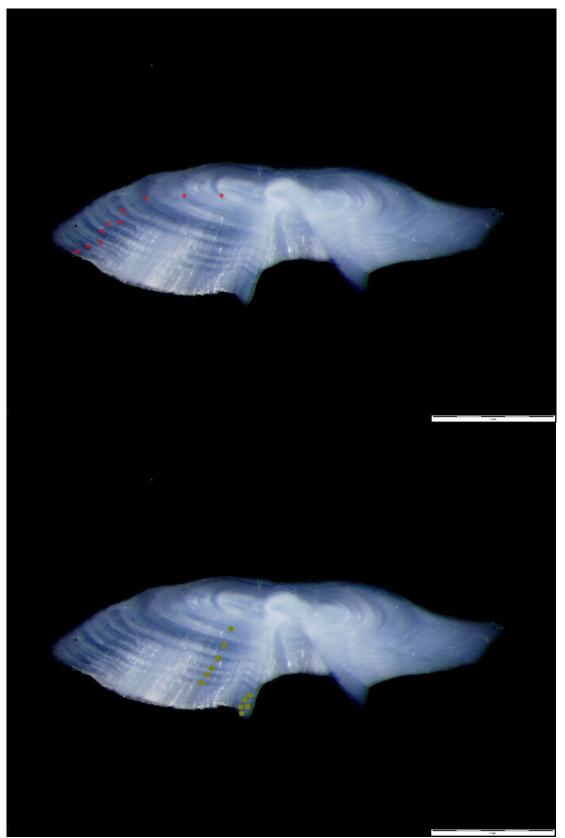


Figure 3. Two different interpretations of an otolith (31.6 cm, January, VII). The agreed interpretation is 12 years old. The first translucent ring close to the nucleus should be counted (as in top photo) and the narrow rings close to the edge should also be counted (as in bottom photo).

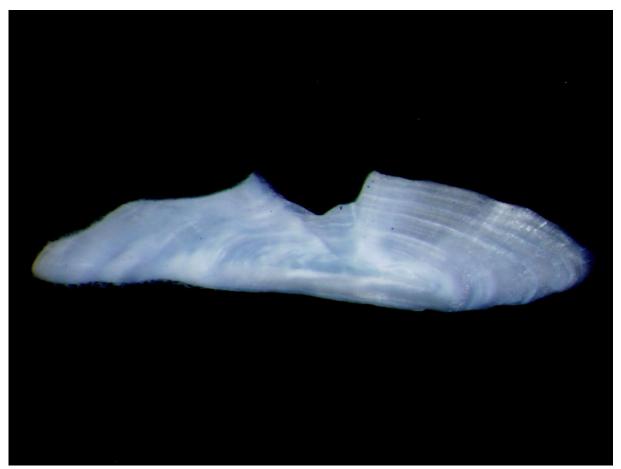


Figure 4. An otolith (29.6 cm, February. VII) with split rings. The agreed interpretation is 6 year old. Some aged this fish to be 7, 8 or 12 years old.

#### 3.3 Workshop results

A subset from set G (subset G1) was included in the exchange. A second subset (G2) was extracted from set G and presented to the readers during the workshop. Both subsets consisted of 4–5 fish per age group in the age range of 4 to 13 ('true' ages), and both subsets were comparable in size distribution (24–35 cm) and catch months (mainly January-March, some from April-May). At the end of the workshop all readers re-read subset G1.

For the sectioned otoliths, the results of the 7 experienced readers showed an improvement from subset G1 to subset G2, and an even greater improvement from subset G2 to the reread of subset G1 (Table 9, Figure 5). Overall agreement with both modal age and 'true' age increased (Table 9). The results by reader for the experienced readers (readers 1-7) showed an increase in agreement from 8-67% to 38-72%, and a decrease in absolute bias from 0-1.8 to 0-0.6 (Table 10). Although the workshop discussions mainly focussed on (images of) sectioned otoliths, the results for broken-burnt otoliths also improved (Tables 9 and 11). It can be argued that the readers may have remembered their first age readings of subset G1, but this seems unlikely because the second reading was carried out 2–7 months later and the readers were not informed that they were reading the same set.

Most of the trainees only participated in the workshop, so subset G2 was the first set for them to read. Comparison of their results for subset G2 and the results of the consecutive age reading for subset G1 showed a tremendous improvement in both accuracy as well as precision. A set of otoliths of young fish was included in the exchange and workshop (NL-VIIe-2003). In the exchange, the agreement with modal age was high (up to 100%) for some readers, but low (down to 13%) for others (Table 12). A new set (consisting of 10 otoliths) and a selection of 10 otoliths from the exchange were re-read at the workshop after the interpretation discussions. A great improvement was observed: the agreement with modal age was 100% for 8 of the workshop participants and 65-95% for the other 4 participants (Table 12). This exercise served as a repetition on how to interpret the first annuli rather than a reliable measurement of accuracy and precision, because the readers were aware that the subsets contained 'more of the same'. The results do however show that consensus on interpretation of the first annuli in young fish was achieved.

Table 9. Overall agreement with modal age and 'true' age for the 7 experienced readers who participated in both the workshop and the exchange (set G).

Otolith act (proparation method)	N	% agr	eement
Otolith set (preparation method)	IN	'true' age	modal age
G1 exchange (sections)	48	43%	53%
G2 w orkshop (sections)	48	48%	56%
G1 w orkshop (sections)	48	58%	62%
G1 exchange (broken-burnt)	48	39%	54%
G1 w orkshop (broken-burnt)	48	48%	56%

Otolith set		Reader 1	Reader 2	Reader 3	Reader 4	Reader 5	Reader 6	Reader 7	Reader 9	Reader 10	Reader 11	Reader 12	Reader 13
G1 exchange	N aged	48	48	48	48	48	48	48	48	0	0	0	0
	Agreement	67%	60%	25%	56%	48%	38%	8%	44%	-	-	-	-
	CV	7%	10%	11%	11%	7%	10%	13%	8%	-	-	-	-
	Bias	0.2	0.0	-1.3	0.3	-0.6	-0.9	-1.8	-0.4	-	-	-	-
G2 w orkshop	N aged	47	48	48	48	48	48	48	48	48	48	48	48
	Agreement	57%	44%	31%	35%	63%	56%	31%	46%	35%	33%	27%	52%
	CV	9%	15%	9%	12%	7%	10%	11%	8%	15%	14%	12%	15%
	Bias	0.1	0.5	0.6	0.8	0.1	0.3	-0.4	0.1	-0.7	0.5	1.0	0.1
G1 w orkshop	N aged	46	48	48	48	48	48	48	48	48	48	48	48
	Agreement	72%	73%	67%	44%	50%	63%	38%	67%	50%	33%	48%	54%
	CV	4%	5%	7%	10%	8%	7%	11%	6%	9%	15%	10%	9%
	Bias	-0.2	0.0	-0.3	0.4	0.3	-0.4	-0.6	-0.1	-0.3	-0.6	0.1	-0.3

Table 10. Sectioned otoliths set G – Comparison with 'true' age.

Table 11. Broken-burnt otoli	hs set G – Com	parison with 'true' a	age.
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Otolith set		Reader 1	Reader 2	Reader 3	Reader 4	Reader 5	Reader 6	Reader 7	Reader 9	Reader 10	Reader 11	Reader 12	Reader 13
G1 exchange	N aged	48	48	48	48	48	48	48	0	0	0	0	0
	Agreement	42%	44%	25%	33%	67%	31%	31%	-	-	-	-	-
	CV	10%	7%	10%	18%	6%	12%	18%	-	-	-	-	-
	Bias	-0.7	-0.6	-1.8	0.2	-0.3	-1.3	-1.2	-	-	-	-	-
G1 w orkshop	N aged	44	48	48	48	47	45	44	0	44	47	0	45
	Agreement	64%	69%	25%	33%	66%	51%	25%	-	39%	26%	-	51%
	CV	8%	6%	10%	18%	9%	11%	15%	-	9%	10%	-	8%
	Bias	-0.5	0.0	-0.9	0.2	-0.2	-0.4	-1.3	-	-0.7	-0.9	-	-0.5

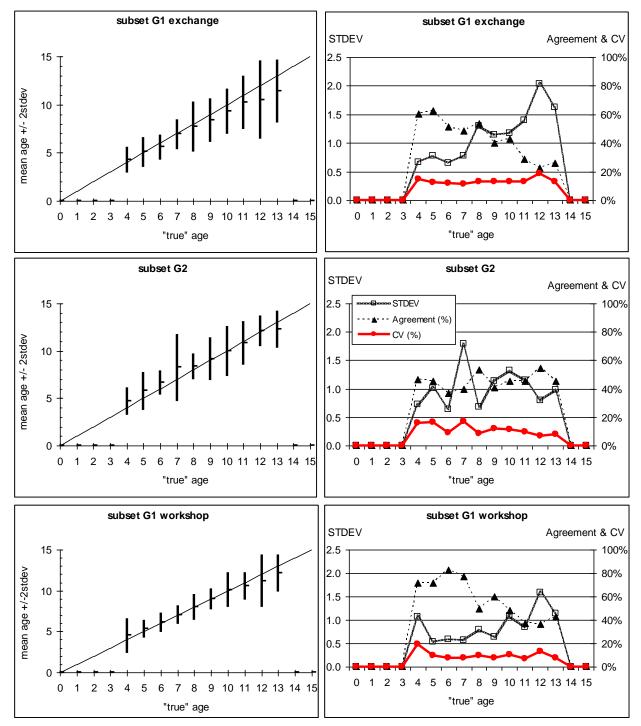


Figure 5. Results of the 7 experienced readers who participated in both the workshop and the exchange. Top panels show the exchange results for subset G1, middle panels show the workshop results for subset G2 (which was designed to be identical to set G1), and bottom panels show the results of the reread of subset G1 during the workshop.

Otolith set		Reader 1	Reader 2	Reader 3	Reader 4	Reader 5	Reader 6	Reader 7	Reader 9	Reader 10	Reader 11	Reader 12	Reader 13
excange	N aged	23	23	23	23	23	23	23	23	0	0	0	0
	Agreement	100%	91%	96%	13%	65%	96%	13%	83%	-	-	-	-
	CV	0%	10%	0%	26%	29%	0%	14%	18%	-	-	-	-
	Bias	0.0	0.1	0.0	-0.9	-0.3	0.0	1.0	-0.1	-	-	-	-
workshop	N aged	20	20	10	20	20	20	20	20	20	10	20	20
	Agreement	100%	100%	100%	100%	100%	100%	75%	100%	90%	100%	65%	95%
	CV	0%	0%	0%	0%	0%	0%	26%	0%	16%	0%	30%	31%
	Bias	0.0	0.0	0.0	0.0	0.0	0.0	-0.2	0.0	-0.1	0	0.5	0.2

Table 12. Otoliths of young fish (set NL-VIIe-2003) – Comparison with modal age.

#### 4. Recommendations

On horse mackerel ageing methods

- Innovative research should be carried out to develop better methods to enhance the contrast between opaque and translucent in sectioned otoliths (especially for Southern stock).
- Although reflected light is the preferred method for reading sections, alternating with transmitted light can sometimes help to interpret the structures.

On workshops

- Frequent workshops should be held for difficult species such as horse mackerel (once every 3–5 years).
- An exchange (shortly) before workshop increases the effectiveness of the workshop.
- Readers attending the workshop should also participate in the exchange.
- Be aware and make clear decisions on how workshop time is allocated over reading vs. discussing images, different areas, different methods, etc.

On training

- Taking trainees to workshops offers an opportunity for a quick start of the learning process.
- The best way to learn is by putting up images on the screen and jointly discussing the interpretation.

On reference collections

• Collate an image collection from the 'known' age set G, in which the agreed interpretation is annotated in a separate layer.

On validation

- In general, calibration alone is not sufficient, validation is also required.
- For horse mackerel, validation of the growth patterns in the first years of life by day-ring analyses. This is not covered (sufficiently) by the 'known' age collections based on an extremely strong year-class.

#### References

- Eltink ATGW, Newton AW, Morgado C, Santamaria MTG, Modin J (2000). Guidelines and tools for age reading comparisons. EFAN Report 3-2000.
- ICES (1999) Report of the Horse Mackerel Otolith Workshop, 15-19 January Lowestoft, UK. ICES CM 1999/G:16
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- ICES (2005) Report of the Working Group on the Assessment of Mackerel, Horse Mackerel, Sardine and Anchovy (WGMHSA), 6 - 15 September, Vigo, Spain. ICES CM 2006/ACFM:08
- ICES (2007) Report of the Planning Group on Commercial Catch, Discards and Biological Sampling (PGCCDBS), 5–9 March 2007, Valetta, Malta. ICES CM 2007/ACFM:09

### Justification

Rapport:	CVO 11.007
Project Number:	4301900335

The scientific quality of this report has been peer reviewed by the a colleague scientist and the head of the department of IMARES.

Approved:	Sieto Verver
	Deputy head of CVO

Signature:

Date: 11 August 2011

Approved:	Jakob Asjes
	Head of department

Signature:

Date: 11 August 2011

#### **Appendix A. Extended Summary**

As published in the report of the Planning Group on Commercial Catch, Discards and Biological Sampling (PGCCDBS) in 2007 (ICES, 2007).

Following a recommendation from PGCCDBS, a workshop on age calibration of horse mackerel was carried out. The last workshop had been held in 1999. Furthermore, the age compositions provided to WGMHSA (Working Group on the Assessment of Mackerel, Horse Mackerel Sardine and Anchovy) and the results of an informal small-scale otolith exchange indicated that ageing problems may exist.

The objectives of the exchange and workshop were:

- a. In general, improve the quality of horse mackerel readings by international calibration.
- b. In particular, attempt to resolve the observed differences between countries.
- c. Estimate the accuracy and precision of the age readings before and after the intercalibration.
- d. Take into account differences between areas and methods.
- e. Training of new horse mackerel readers.

The exchange and the workshop were carried out in 2006. Eight experienced readers participated in the exchange, 7 of which also participated in the workshop. Five trainees participated in the workshop, only one of them also participated in the exchange. All countries providing age reading data to the WGMHSA were represented in both the exchange and the workshop by an experienced reader.

Portugal, Germany and The Netherlands provided otolith sets for the exchange. The sets represented different otolith preparation methods and stocks (Table A1). Sets G & K consisted of otoliths from the extremely strong 1982 year-class and hence the age is considered to be known (with a certainty of approximately 95%). Set NL-VIIe-2003 focused on the young fish, as these were expected to present problems based on the informal small-scale otolith exchange.

The experienced readers were accustomed to different otolith preparation methods and different growth patterns associated with the different stocks. Generally, the readers had more difficulty if they were reading material they were not accustomed to.

Horse mackerel is regarded to be a difficult species to age and this is reflected by the results of the exchange (Table A1). The agreement between the experienced readers was low, especially for otoliths from the Southern stock. For sets G and K, the agreement with modal age was higher than with 'true' age. Comparison with the 'true' ages showed an overall tendency to underestimate the age (top left panel in Figure 1A).

Otalith act (proportion mathed)	Stock	Months	Cine renge	A	Number	of otoliths	% agreement	
Otolith set (preparation method)	set (preparation method) Stock Months Size range Age range		Age range -	total	exchange	'true' age	modal age	
PT-IXa-2005 (broken-burnt/w hole)	Southern	1-3	21-42 cm	2-21	51	51	n.a.	38%
PT-IXa-2005 (sections)		"		2-24	51	50	n.a.	37%
DE-IV-2005 (sections)	North Sea	7-8	19-33 cm	1-13	175	48	n.a.	62%
DE-VIId-2005 (sections)	Western	10	18-37 cm	1-21	233	51	n.a.	58%
NL-VIIe-2003 (sections)	Western	8	16-20 cm	1-4	100	23	n.a.	68%
G (broken-burnt)	Western	1-5	24-35 cm	4-13	170	48	39%	54%
G (sections)		"	"	3-15	170	48	43%	53%
K (sections)	Western	9-11	14-30 cm	1-12	153	38	36%	56%

Table A1. Description of the otolith sets included in exchange and percentage agreement for the 7 experienced readers who participated in both the exchange and the workshop.

For the Dutch sets (set G, set K and set NL-VIIe-2003), digital images of the sectioned otoliths were annotated by the readers participating in the exchange. During the workshop these annotated images were used to discuss differences in interpretation. A great deal of attention was paid to the interpretation of the first annuli, both in young fish as well as in older fish. This point appeared to be the major cause of differences in interpretation. In some otoliths split rings or the interpretation of the edge of the otoliths caused problems. All these features were discussed and eventually consensus was reached for all otoliths put up on the screen.

For a small set of the Southern stock otoliths provided by Portugal, images of sectioned otoliths were digitised during the meeting. These images were discussed in the group. In some cases consensus could be reached on how to interpret the otolith, however in other cases it seemed to be impossible to age the otolith. Ageing of the Southern stock otoliths appeared to be less difficult when using broken-burnt material instead of (images of) sectioned otoliths.

A subset was extracted from set G (subset G1) and included in the exchange. A second subset (G2) was extracted from set G and presented to the readers during the workshop. Both subsets consisted of 4–5 fish per age group in the age range of 4 to 13 ('true' ages), and both subsets were comparable in size distribution (24–35 cm) and catch months (mainly January-March, some from April-May). At the end of the workshop all readers re-read subset G1. The results of the 7 experienced readers clearly showed an improvement from subset G1 to subset G2, and from subset G2 to the reread of subset G1 (Figure A1). Although it can be argued that the readers may have remembered their first age readings of subset G1, this seems unlikely because the second reading was carried out 2–7 months later and the readers were not informed that they were reading the same set.

A similar select, re-select, and reread of first selection was carried out for 2 small subsets from the set containing only young fish (NL-VIIe-2003). The percentage agreement increased to almost 100%. However, this exercise served more as a repetition on how to interpret the first annuli than as a reliable measurement of accuracy and precision because the readers were aware that the subsets contained 'more of the same'.

Most of the trainees only participated in the workshop, so subset G2 was the first set for them to read. Comparison of their results for subset G2 and the results of the consecutive age reading for subset G1 showed a tremendous improvement in both accuracy as well as precision.

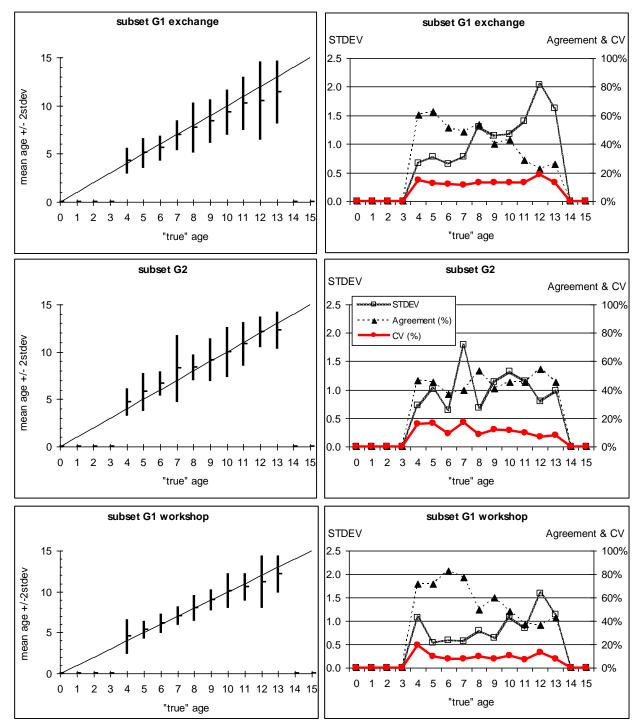


Figure A1. Results of the 7 experienced readers who participated in both the workshop and the exchange. Top panels show the exchange results for subset G1, middle panels show the workshop results for subset G2 (which was designed to be identical to set G1), and bottom panels show the results of the reread of subset G1 during the workshop.

#### Recommendations

On horse mackerel ageing methods

- Innovative research should be carried out to develop better methods to enhance the contrast between opaque and translucent in sectioned otoliths (especially for Southern stock).
- Although reflected light is the preferred method for reading sections, alternating with transmitted light can sometimes help to interpret the structures.

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• Collate an image collection from the "known" age set G, in which the agreed interpretation is annotated in a separate layer.

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- In general, calibration alone is not sufficient, validation is also required.
- For horse mackerel, validation of the growth patterns in the first years of life by day-ring analyses. This is not covered (sufficiently) by the "known" age collections based on an extremely strong year-class.

Reader	Expertise	Exchange	Workshop	Country	Name
1	experienced	yes	yes	DE	Gudrun Gentschow
2	experienced	yes	yes	NO	Helga Gill
3	experienced	yes	yes	DK	Aage Thaarup
4	experienced	yes	yes	NL	Simon Rijs
5	experienced	yes	yes	IR	Eugene Mullins
6	experienced	yes	yes	ES	lñaki Rico
7	experienced	yes	yes	PT	Ana Moreira
8	experienced	yes	no	PT	Maria João Ferreira
9	trainee	yes	yes	NL	André Dijkman
10	trainee	no	yes	DE	Cornelia Albrecht
11	trainee	no	yes	DK	Gert Holst
12	trainee	no	yes	IR	Turloch Smith
13	trainee	no	yes	ES	Clara Dueñas

## Appendix B. Age Readers ID