



# RELATÓRIOS CIENTÍFICOS E TÉCNICOS

## SÉRIE DIGITAL

**REPORT OF THE WORKSHOP ON SARDINE OTOLITH  
AGE READING (LISBON, 28 JANUARY - 1 FEBRUARY, 2002)**

**Eduardo Soares, Alexandre Morais, Alexandra Silva, Pablo Carrera,  
Afonso Jorge, Iñaki Rico, Quena Peleteiro and Hughes Evano**

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# REPORT OF THE WORKSHOP ON SARDINE OTOLITH AGE READING (LISBON, 28 JANUARY – 1 FEBRUARY, 2002)

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## ABSTRACT

An otolith exchange for sardine was carried out in July 2000 within the framework of EU Project PELASSES to standardise age reading criteria between project participants. A total of 359 otolith pairs were analysed from sardine samples collected in the spring acoustic surveys covering the area from the English Channel to the Gulf of Cadiz. Disagreement in age readings of young (age groups 1 and 2) and old fish (from age group 4 onwards) and on otoliths from the southern areas (Algarve and Cadiz) were the main problems identified during the exchange and later discussed on a workshop. The consistency within readers was also checked during the workshop. Identification of the first annual ring was the main problem on younger ages. In older fish, discrimination of rings near the otolith edge caused most of the disagreements. These difficulties are complicated in otoliths from the southern areas, due to the less clear structure and to the frequency of occurrence of false rings. A poor consistency within readers was also observed. The present workshop outlined an improvement in sardine age reading performance since the last otolith exchange with acceptable levels of agreement, precision and accuracy for young individuals (age groups one to three). However, ageing older individuals, otoliths from the southern area and within reader consistency are still a matter of concern.

**Keywords:** sardine, age reading, intercalibration, otoliths

## RESUMO

**Título: Relatório do Workshop Sobre Leitura de Idades em Otólitos de Sardinha (Lisboa 28 de Janeiro – 1 de Fevereiro, 2002)** Em Julho de 2000, realizou-se um intercâmbio de otólitos de sardinha no âmbito do Projecto PELASSES da UE com o objectivo de uniformizar os critérios de determinação de idades entre os participantes do projecto. Foram analisados 359 pares de otólitos recolhidos nas campanhas acústicas realizadas na Primavera e cobrindo a área desde o Canal da Mancha até ao Golfo de Cádiz. Os principais problemas identificados relacionam-se com a determinação de idades dos indivíduos jovens (grupos de idade 1 e 2) e velhos (a partir da idade 4) e com os otólitos das áreas do sul (Algarve e Cádiz). Estes problemas e ainda a consistência de cada leitor foram discutidos mais tarde num *workshop*. A identificação do primeiro anel anual é a principal causa de discordância nas idades jovens. Nos peixes mais velhos, a discriminação dos anéis junto ao bordo do otólito é uma das dificuldades principais. Estas dificuldades são agravadas nos otólitos da zona sul, devido à sua estrutura menos clara e à frequência de anéis falsos. Constatou-se também uma baixa consistência intra-leitor. Este workshop salientou uma melhoria da determinação de idades da sardinha em relação ao exercício anterior tendo sido atingidos níveis aceitáveis de concordância, precisão e rigor para os indivíduos jovens (idades 1 a 3 anos). No entanto, a determinação da idade dos indivíduos mais velhos ou provenientes da zona sul e a consistência dos leitores são ainda motivos de preocupação.

**Palavras-chave:** sardinha, determinação de idades, intercalibração, otólitos.

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## REFERÊNCIA BIBLIOGRÁFICA

SOARES, E.; MORAIS, A.; SILVA, A., CARRERA, P.; JORGE, A.; RICO, I.; PELETEIRO, Q.; EVANO, H. 2002. Report of the workshop on sardine otolith age reading (Lisbon, 28 January – 1 February, 2002). *Relat. Cient. Téc. IPIMAR, Serie digital* (<http://ipimar-iniap.ipimar.pt>) nº 14, 26p.

## INTRODUCTION

An otolith exchange for sardine was carried out in June-July 2000 with the general aim of standardising age reading criteria between the participants of Project PELASSES (EU DG FISH, Project no. 99/010). Particular attention was given to the comparison of sardine otoliths from different locations within the project study area (from the Celtic Sea to Gibraltar).

The exchange was based on a sample of otoliths from different areas collected during the project surveys in spring 2000 and involved six readers of the three partner countries (Portugal, Spain and France). Several difficulties were identified during the exchange and were later discussed on a Workshop that took place in the Instituto Nacional de Investigação Agrária e das Pescas - INIAP/IPIMAR (Lisbon, 28 January-1 February 2002), with the main objectives:

1. To discuss age reading differences in young and old fish and establish criteria to improve the precision of age readings;
2. To identify the main difficulties in age readings of otoliths from southern areas and establish new criteria to improve ageing agreement;
3. To check consistency within readers;

## Participants

Name	Institute	Exchange	Workshop
Eduardo Soares ( <b>ES</b> ) (Chairman)	INIAP/IPIMAR	X	X
Afonso Jorge ( <b>AJ</b> )	INIAP/IPIMAR	X	X
Pablo Carrera ( <b>PC</b> )	IEO	X	X
Quena Peleteiro ( <b>QP</b> )	IEO	X	X
Evano Hugues ( <b>EV</b> ) (new reader)	IFREMER	X	-
Iñaki Rico ( <b>IR</b> ) (new reader)	AZTI	X	X
Raquel Milhazes (observer)	INIAP/IPIMAR	-	X
Alexandre Morais (observer)	INIAP/IPIMAR	-	X
Delfina Morais (observer)	INIAP/IPIMAR	-	X
Lurdes Pires (observer)	INIAP/IPIMAR	-	X
Susana Rodrigues (observer)	INIAP/IPIMAR	-	X
Alexandra Silva (observer)	INIAP/IPIMAR	-	X

The first three readers are responsible for the preparation of sardine age-length keys from surveys and commercial catches in each country. IR has experience in reading other species otoliths, namely anchovy, but no experience on sardine and EV has no experience in age reading. The workshop was an opportunity to train new otolith readers and six other persons participated as observers.

## MATERIAL AND METHODS

### Exchange

A protocol for the exchange was prepared and circulated among the participants. A total of 359 otolith pairs were analysed from sardine samples collected in the Celtic Sea and off the coasts of France, Spain and Portugal during surveys carried out in Spring 2000 (Table 1). Otoliths were prepared according to the procedure described in the 1997 Workshop on Sardine Otolith Age Reading and age determination followed the protocol adopted in that Workshop (ICES, 1997).

Table 1-Geographic distribution of the otolith samples.

ICES Division	Area	Length range (cm)	No. of otoliths	
			Exchange	Workshop
VII	British Channel	12.5 - 26.0	30	6
VII	Brittany	12.5 - 25.5	50	14
VIII	Gulf of Biscay	16.0 - 21.5	20	7
VIII	Basque Country	16.5 - 24.5	31	13
VIII	Cantabria	18.0 - 23.5	23	8
VIII	Asturias	18.5 - 25.0	24	-
VIII	North Galicia	20.5 - 23.5	14	-
IXa N	South Galicia	15.0 - 21.0	23	-
IXa N	Oporto	11.0 - 22.0	45	11
IXa S	Lisbon	11.0 - 19.5	37	7
IXa S	Algarve	16.0 - 23.0	31	9
IXa S	Cadiz	14.0 - 21.5	31	5
	Total	11.0 - 26.0	359	80

Each reader classified the otoliths according to a scale of readability (three levels: good, medium and difficult), interpreted the type of edge (hyaline-H, opaque-O) and counted the number of hyaline rings. Percentage readability and edge type were calculated for each sample and reader and averaged across all readers. The birthdate was assumed to be 1 January and the hyaline edge was included in the age. For example, a fish was classified into age group 2 if the otolith had one hyaline ring and a hyaline edge or if it had two hyaline rings and an opaque edge. Age readings for the whole collection of otoliths were compared among readers and between readers and the modal age. Agreement among readers is compared with the results of the 1997 Workshop. Data from the areas covered by the Spanish survey (areas VII and VIII) and by the Portuguese survey (area IXa-N and IXa-S) were analysed separately,

mainly to investigate if there are differences in age determinations between the regular readers of each survey and the non regular ones. The mean differences between readers in age determination for each (modal) age group were computed for each area.

## **Workshop**

A sub-sample of 80 otolith pairs from the exchange, covering the different age groups and areas, was re-read at the beginning of the workshop in order to test the consistency of each reader (Table 1). Age determinations from the exchange (excluding the reader EV and only taking into account the 80 otolith pairs) were considered as the first reading and those of the workshop as the second reading.

Two sets of 24 and 25 otoliths from young (less than 3 year old) and old (more than 5 year old) fish with a high consensus (more than 80% agreement between all readers) were reviewed and discussed in a group in order to calibrate age reading criteria. Subsequently two sets of young and old otoliths showing poor agreement in the exchange (less than 70%) were analysed and discussed by all readers. A similar procedure was followed with a sample of 50 otoliths from the southern area (Algarve and Cadiz).

The sub-sample of 80 otolith pairs was re-read at the end of the workshop (third reading) after the discussion of the difficulties and the agreement of additional criteria, to evaluate improvement in the ageing consistency between readers.

An image analysis system was used in group discussions. All data were analysed using the workbook for age reading comparisons (Eltink, 2000) and following the recommendation of the guidelines and tools for age reading comparisons (Eltink *et al.*, 2000).

## **RESULTS AND DISCUSSION**

### **Otoliths exchange**

#### Readability of the otoliths

Classification of otoliths from each sample was variable among readers (criteria for each level were not defined and the quality of preparations deteriorated along the circulation) but on the average, more than 75% of the otoliths were considered with good or average readability (Table 2). The highest percentages of otoliths with good readability were observed in the

northern areas, mainly in samples from the British Channel, Brittany, the Gulf of Biscay, the Basque Country, Cantabric Sea and Oporto. Difficult otoliths were found in the Southern area (Algarve and Cadiz) but also in South Galicia and Lisbon.

Table 2 – Average readability of sardine otoliths for each sample.

Area	Readability (%)		
	Good	Medium	Difficult
British Channel	83	13	4
Brittany	87	11	2
Gulf of Biscay	76	19	5
Basque Country	74	23	3
Cantabrian Sea	81	12	6
Asturias	64	34	2
North Galicia	60	33	7
South Galicia	57	27	16
Oporto	78	14	7
Lisbon	57	26	17
Algarve	34	45	21
Cadiz	28	48	25

### Otolith edge

Agreement among readers in the interpretation of the edge was generally high. The majority (>86%) of otolith edges were hyaline in each area as expected for this time of the year (March/April), except in the sample from Lisbon where 60% were opaque corresponding to small fishes (12.5-14 cm) (Table 3). Opaque edges were observed in lower percentages in Oporto (14%) and Britany (12%).

Table 3 – Average percentage of otolith edge type for each sample.

Area	Edge type (%)	
	Hyaline	Opaque
British Channel	99	1
Brittany	88	12
Gulf of Biscay	97	3
Basque Country	98	2
Cantabrian Sea	98	2
Asturias	98	2
North Galicia	99	1
South Galicia	98	2
Oporto	86	14
Lisbon	40	60
Algarve	92	8
Cadiz	92	8

#### Age readings

The average percentage of agreement with the modal age across all ages and readers in the exchange was 76.1% and the average CV was 15.5% (Annex 1). Agreement between readers ranged from 53% to 72% (all ages) and improved marginally when only age groups 1-7 were considered (54-73%) since most of the otoliths (96%) were aged up to 7 years old (Table 4). AJ (experienced reader) and IR (no experience on sardine) have the best agreement (73%), followed by PC with EV (70%) and with the former two readers (68%). Both QP and ES have better agreement with readers from their own countries. These results represent an improvement since the last otolith Workshop (ICES, 1997) when for the same readers, agreement ranged from 31% to 68% (for ages 1-7). The lower values of the range, corresponding to comparisons with QP and ES increased particularly. As in the 1997 Workshop, most of the differences in age readings are of one year (23-35% for ages 1-7, Table 5).

The overall mean CV (using modal age as a reference) was 20% with larger values on the youngest age groups (25% on age 1 and 24% on age 2) (Table 6). The average CV ranged from 16% (AJ) to 25% (ES and QP) and most readers tend to be less precise in younger ages. However the pattern of precision with age is variable among readers. Mean agreement (all readers) with the modal age decreased from 92% at age 1 to 46% at age 7) and was maximum for AJ (84%) and minimum for QP and EV (60%) (Table 7).

Table 4 – Percentage of agreement among readers; for all age groups in upper diagonal, for ages 1 to 7 in lower diagonal and with modal age (ages 1-7) in the diagonal.

	AJ Reader 1	IR Reader 2	PC Reader 3	ES Reader 4	QP Reader 5	EV Reader 6
Reader 1	<b>87</b>	72	67	63	60	65
Reader 2	73	<b>80</b>	66	60	58	65
Reader 3	68	68	<b>79</b>	59	64	68
Reader 4	64	60	61	<b>72</b>	53	59
Reader 5	61	59	66	54	<b>69</b>	64
Reader 6	67	67	70	60	66	<b>77</b>

Table 5 – Percentage of disagreement of one year (upper diagonal) and of two or more years (lower diagonal) among readers.

	AJ Reader 1	IR Reader 2	PC Reader 3	ES Reader 4	QP Reader 5	EV Reader 6
Reader 1	-	23	26	28	32	25
Reader 2	4	-	27	33	32	24
Reader 3	6	5	-	32	24	22
Reader 4	8	6	7	-	35	28
Reader 5	6	9	10	11	-	29
Reader 6	8	8	8	11	5	-

Table 6 – Coefficient of variation (%) of age readings by modal age and reader.

Modal Age	AJ Reader 1	IR Reader 2	PC Reader 3	ES Reader 4	QP Reader 5	EV Reader 6	Mean All Readers
1	31	30	15	37	36	0	25
2	23	12	25	33	34	18	24
3	17	16	17	23	21	25	20
4	9	18	16	28	15	15	17
5	14	22	15	23	27	20	20
6	6	20	14	15	29	20	17
7	16	14	24	19	13	21	18
8	6	7	19	12	13	14	12
9	-	-	-	-	-	-	-
10	-	-	-	-	-	-	-
Mean (1-7)	16	19	18	25	25	17	20

Table 7 – Percentage of agreement with modal age by reader and modal age.

Modal Age	AJ Reader 1	IR Reader 2	PC Reader 3	ES Reader 4	QP Reader 5	EV Reader 6	Mean All Readers
1	89	87	98	88	89	100	92
2	89	94	77	68	52	88	78
3	83	77	79	68	70	60	73
4	87	72	70	45	64	58	66
5	90	60	45	40	65	55	59
6	89	63	50	70	48	33	59
7	58	33	50	75	33	25	46
8	75	75	33	50	33	25	49
9	0	0	100	0	100	0	33
10	100	0	0	0	100	0	33
Mean (1-7)	84	69	67	65	60	60	67

Relative bias to modal age is not a serious problem in age groups 1 to 4, however four of the readers tend to underestimate the older ages (Fig. 1). The average differences from modal age were in the range 0.2 years for age 1 but ranged from  $-0.35$  years to  $+1$  year at age 6.

#### Area disaggregated age readings: Spanish survey and Portuguese survey

When data is disaggregated according to the ICES areas covered by the Spanish (VII and VIII) and Portuguese surveys (IXa-N and IXa-S), the sample size becomes small mainly from age three onwards. Otoliths from Division IXa-S (Algarve+Cadiz) show less precise readings and lower agreement with modal age, clearly presenting more difficulties than otoliths from other areas (Tables 8 and 9). Age readings from area VII (new for all the readers) didn't raise special problems providing CV's and levels of agreement comparable to those from area IXa-N.

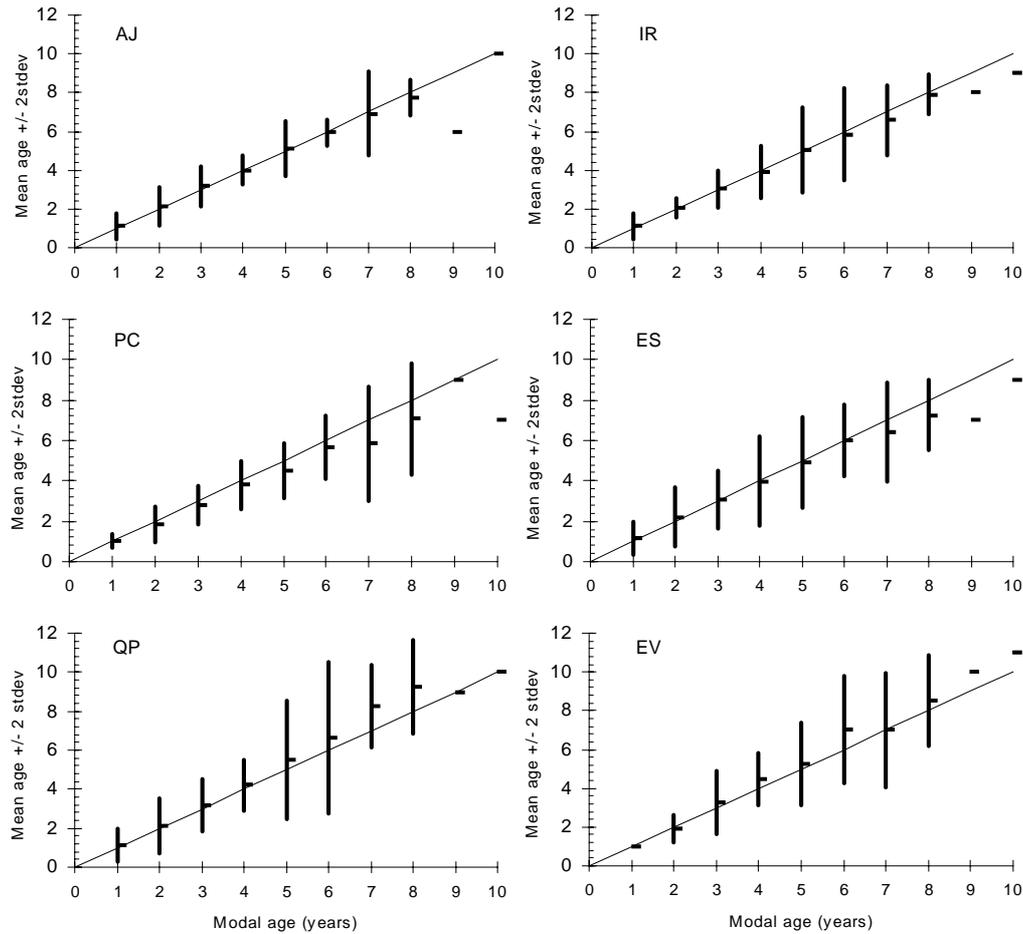


Figure 1 – Age bias plots: mean age +/- 2 standard deviations of each reader against the modal age. The solid line represents the 1:1 relationship between modal age and mean age.

Table 8 – Percentage of agreement with modal age by area, for all readers.

Modal Age	VII	VIII	IXaN	IXaS
1	94	89	94	74
2	81	85	73	64
3	76	81	63	68
4	66	70	62	42
5	58	60	83	55
6	56	61	71	55
7	100	42	-	40
8	47	50	-	50
9	-	33	-	-
10	33	-	-	-
Mean (1-7)	76	70	74	57

Agreement among readers and between readers and the modal age, was generally higher for the Spanish survey (52-79% between readers and 57-84% with modal age) than for the Portuguese survey (53-71% between readers and 41-82% with modal age). Mean differences in age are generally lower in the northern area and bias is not noticeable except in comparisons with QP, that tends to overestimate age (mainly older ages) in relation to the other readers (Fig. 2). In the southern area, AJ tends to give more age than the other readers (particularly PC) and PC tends to give less age than QP and ES (negligible in age 1) (Fig. 3). These results suggest that otoliths from the southern area would be given lower ages if read by PC (who regularly reads otoliths from the northern area).

Table 9 – Coefficient of variation (%) of age readings by modal age and area, for all readers.

<b>Modal Age</b>	<b>VII</b>	<b>VIII</b>	<b>IXaN</b>	<b>IXaS</b>
1	11	18	8	35
2	21	13	23	29
3	13	13	20	18
4	12	13	17	33
5	11	14	8	19
6	19	16	9	15
7	0	18	-	20
8	16	13	-	13
9	-	18	-	-
10	15	-	-	-
<b>Mean (1-7)</b>	<b>12</b>	<b>15</b>	<b>14</b>	<b>24</b>

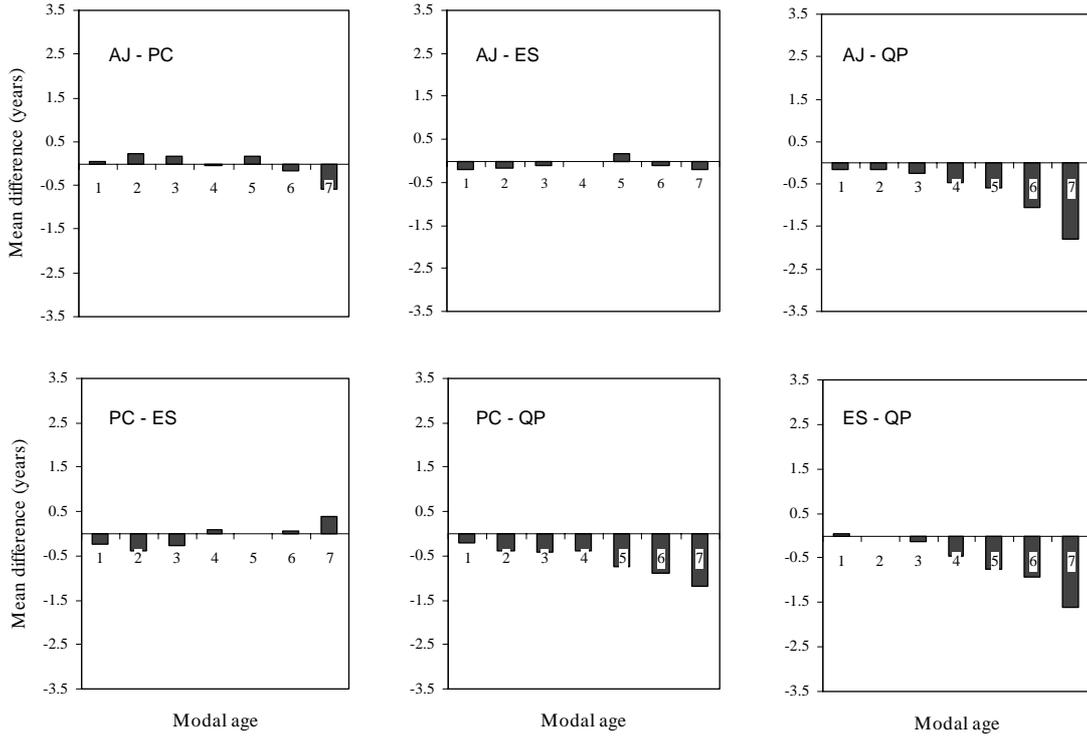


Figure 2 – Mean differences (by modal age) between age readings for each pair of readers, in the Spanish survey.

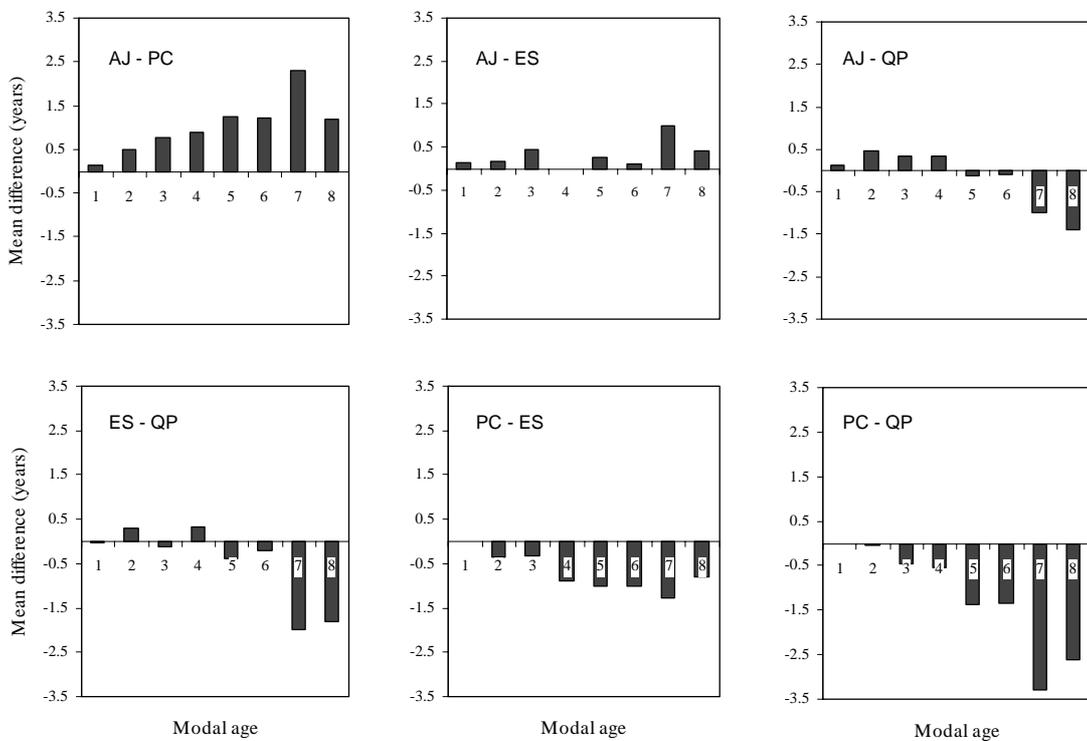


Figure 3 – Mean differences (by modal age) between age readings for each pair of readers, in the Portuguese survey.

## **Workshop**

### Discussion of age determination difficulties: young and old fish and the southern areas

Discussion of the sub-sets of consensual otoliths outlined the similarity between the ageing criteria used by the different readers and was also a means to improve uniformity. Most of the differences in ageing young individuals were due to the identification of the first annual ring. Several false rings appear close to the nucleus and the distance to the first true age ring is variable due to the extended spawning season. The measurement of the ring radius or diameter and its comparison with an average value for the cohort was considered a useful auxiliary criterium. The pattern of ring deposition, reflecting otolith growth, was also considered a helpful observation since a pattern of initial small (or large) distances between rings will likely continue throughout fish growth and a very clear ring structure is generally common to the whole otolith.

Hyaline rings become increasingly narrow and closely packed as fish grow, particularly in a small otolith like that of sardine. Therefore, the discrimination and counting of rings was identified as the main difficulty in ageing older individuals. The use of a higher magnification to observe the otolith edge was an obvious suggestion to reduce the difficulties.

The structure of otoliths from the southern areas is less clear showing several hyaline rings of similar appearance from which is impossible to distinguish true annual rings. False rings are more frequent in the antirostrum, so the participants agreed to use the rostrum for age determination.

### Consistency within readers

A poor percentage of agreement was noticed between the first and second age reading, ranging from 46 to 63% for the different readers (Table 12). The most consistent readers were IR (63%) and PC (61%) and agreement globally decreases with age being above 75% only on the first two age groups. In general there are no signs of bias, except for PC and QP, the former tending to assign older ages on the second reading and the latter showing the opposite trend.

Some of the readers mentioned that the use of a stereomicroscope and light source different from those they are used to could explain part of the differences in the age readings. However,

it seems that the difficulties with the older individuals are the main responsible for the observed poor consistency.

Table 12 – Percentage of reading agreement and bias test within each reader.

	1	2	3	4	5	6	7	8	9	10	Total	
AJ	79	67	50	44	40	70	17	0	0	-	51.3	(-)
IR	90	75	75	77	33	56	20	50	25	0	62.5	(-)
PC	86	80	75	50	42	50	56	20	33	0	60.0	(**)
ES	77	83	38	33	38	55	43	40	0	-	53.2	(-)
QP	84	54	13	30	100	43	33	33	33	0	45.6	(**)

-	= no sign of bias ( $p > 0.05$ )
*	= possibility of bias ( $0.01 < p < 0.05$ )
**	= certainty of bias ( $p < 0.01$ )

### Third reading

The sub-sample of 80 otoliths used in the workshop was not randomly selected from the exchange sample (one of the differences being a lower representation of the youngest ages on the former), so that their results are not comparable. The third reading was therefore contrasted with the results from the exchange for the same otolith sub-sample (first reading).

Results from the third reading show an improvement both of precision and of agreement (around 10%) with the modal age for age groups 2 and 3 and one particular reader (QP) globally increasing age reading performance (Fig. 4, Table 13). Results for other age groups and readers were similar to the first reading, therefore the average percentage of agreement and CV across all ages and readers does not represent an improvement (75% and 12% against 73% and 14%, respectively).

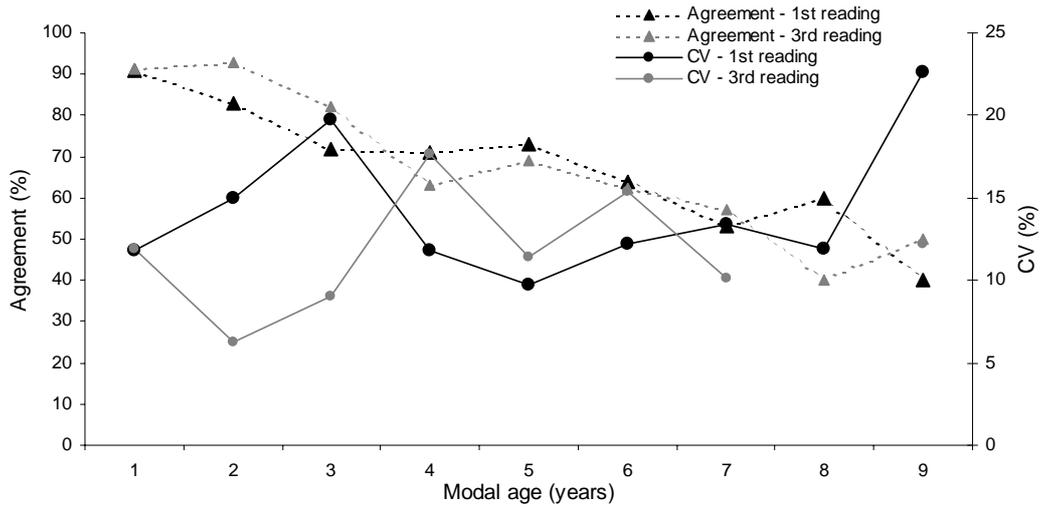


Figure 4 – Coefficient of variation (CV%) and percentage of agreement against the modal age for the 1<sup>st</sup> and 3<sup>rd</sup> reading.

Table 13 – Percentage of agreement among readers; for all age groups and inter-reader bias test (upper diagonal -1<sup>st</sup> reading, lower diagonal - 3<sup>rd</sup> reading) and reader against modal age bias test.

	AJ Reader 1	IR Reader 2	PC Reader 3	ES Reader 4	QP Reader 5
Reader 1	-	64.6 (-)	64.6 (*)	60.8 (-)	53.2 (**)
Reader 2	65.0 (-)	-	57.0 (-)	50.0 (-)	55.0 (**)
Reader 3	62.5 (-)	66.3 (-)	-	53.2 (-)	54.4 (**)
Reader 4	64.1 (**)	48.7 (*)	53.8 (**)	-	43.8 (**)
Reader 5	63.8 (-)	65.0 (-)	53.8 (-)	56.4 (*)	-
Modal age (1 <sup>st</sup> reading)	-	-	-	-	**
Modal age (3 <sup>rd</sup> reading)	-	-	-	**	-

-	= no sign of bias ( $p > 0.05$ )
*	= possibility of bias ( $0.01 < p < 0.05$ )
**	= certainty of bias ( $p < 0.01$ )

The bias trend in older ages (from age four onwards) persisted in the third reading showing a decrease for all readers and particularly for QP. However, PC maintained the bias level but with an opposite sign (positive) and ES increased underestimation tendency. (Fig. 5).

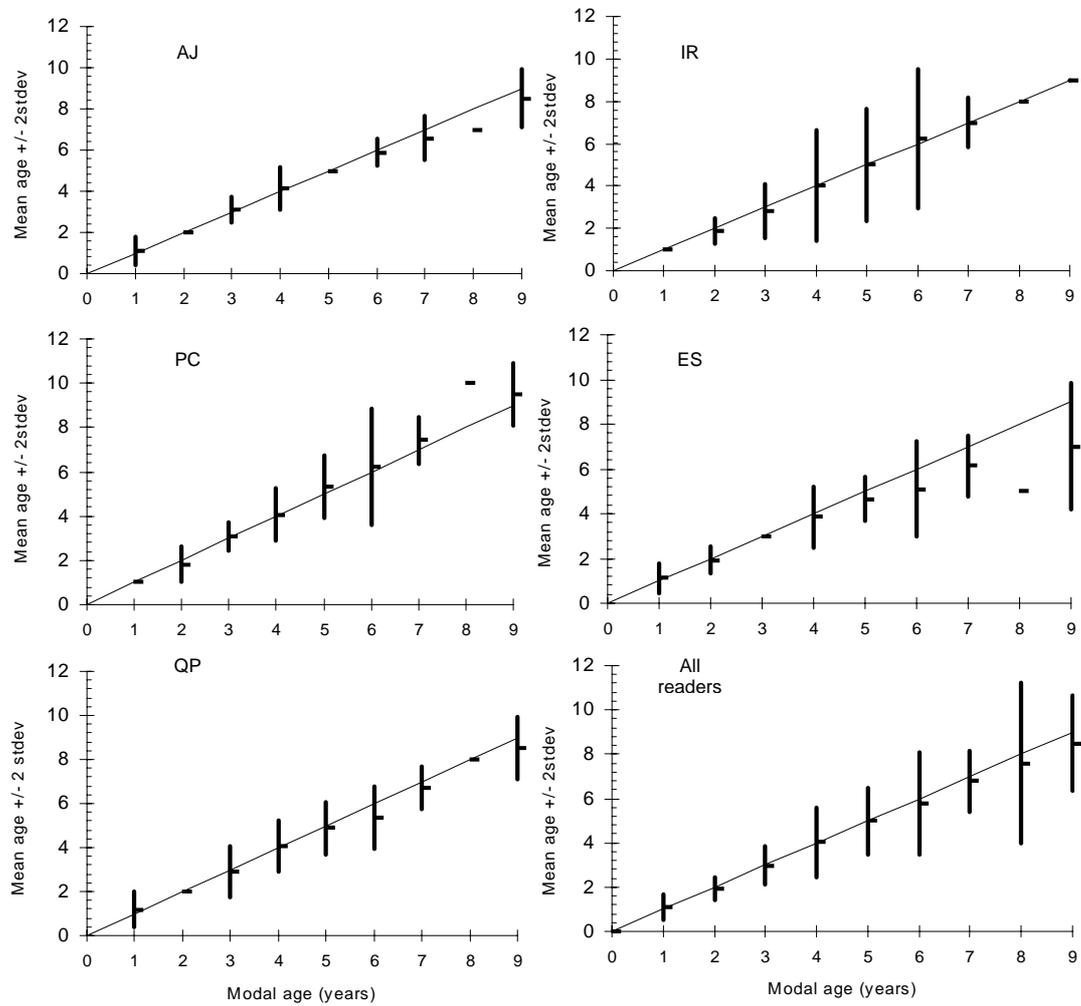


Figure 5 – Age bias plots: mean age +/- 2 standard deviations of each reader against the modal age. The solid line represents the 1:1 relationship between modal age and mean age.

## **AGE READING OF OTOLITHS FROM MEDITERRANEAN, MOROCCO AND AZORES AREAS**

During the workshop a small sample of sardine otoliths (53 pairs) mostly from young fishes (ages 1 and 2), from Mediterranean, Morocco and Azores were observed in order to detect any reading difficulties on otoliths from other areas. In general there was a good agreement among the readers (93% of agreement with the modal age and a CV of 5.5%), showing that these otoliths did not present major difficulties.

## **CONTRIBUTION TO THE WORKSHOP**

As a contribution to this workshop the main results of the “Sardine Otoliths Age Reading Workshop” which took place in Kaliningrad (Russia), 28-31 August 2001, within the FAO–Nansen project (GCP/INT/730/NOR) and RIVO, were presented by Cristina Morgado from INIAP/IPIMAR, who attended that workshop. Moroccan sardine otoliths were discussed by readers from Spain, Morocco and Russia.

Several age reading criteria were agreed by these countries in order to improve the precision of their age readings: The 1st of January was considered as the date of birth. After the nucleus each set of opaque and translucent rings are considered as 1 year. False rings are narrower than annual rings. At rostrum only annual rings are observed. Translucent edge in 1st quarter is considered as an annual ring. Opaque zone at the edge is not considered except in January-February, when this zone is comparable to the annual ring in width. Concerning first annual ring location it was agreed that it should be based on quantitative measurements based on otolith microstructure research.

Several recommendations were agreed in Kaliningrad. Concerning the methodology it was agreed that among other criteria, the reliability of the age reading should be taken into account and that it would be convenient that 2 readers from each country could read the otoliths, discuss and agreed with the age readings from their country. Concerning reading criteria, it was recommended that an exchange programme of 50 otoliths should be carried out between countries on an annual basis. Ring measurements of some exchange otoliths (e.g. 50) should be taken, in order to check if all readers are considering the same rings. If the results of the exchange show low precision in the age readings a workshop should be carried out. The work on daily increments should be encouraged, in order to improve the knowledge of sardine age

estimation. Finally, it was recommended that a guideline for age readings be prepared by FAO /FIRM.

## **CONCLUSIONS AND RECOMMENDATIONS**

The present workshop outlined an improvement in sardine age reading agreement and precision since the last otolith exchange (ICES, 1997), particularly for the readers who were previously more divergent. An acceptable level of agreement (above 80%), precision (CV around 10%) and no sign of bias relative to the modal age was achieved for young individuals (age groups one to three), a reassuring result since these age groups often dominate survey and fishery catches (ICES, 2002).

Identification of the first annual ring is the main justification for disagreement in young age groups and studies of first ring measurements and edge type along the year are recommended to help clarify this question. Divergences in ageing older individuals still persist although a decrease in bias was noticed for most of the readers. The difficulty to discriminate and count narrow closely packed rings near the otolith edge explains the disagreements, which are hard to eliminate because they are linked to the skills of each reader. However, the participants agreed that the use of a higher magnification near the otolith edge could be beneficial (contrary to the current guideline of a low amplification common to the whole otolith).

The highest percentages of agreement, ranging from 68 to 73%, were observed among readers AJ, IR, PC and EV. The two readers with no previous experience on sardine (IR and EV) had levels of agreement, precision and bias (relative to the modal age), comparable to those of the regular readers. A poor consistency within readers was observed, the percentages of agreement across all ages being lower within readers (45-63%) than among readers. Although the main problems are once again on older ages, the coherence of sardine age readings becomes a matter of concern. To minimise this problem, each reader should regularly calibrate his age readings with a set of otoliths with high agreement among readers. It is recommended that such a reference collection be prepared from different seasons (e.g. quarters) and areas.

Otoliths from the southern area (Algarve and Cadiz) present more problems of readability, agreement and age reading precision, as already noted in the 1997 exchange, while otoliths from ICES sub-area VII (which were analysed on a workshop for the first time) did not raise

particular problems. The observed bias between Portuguese and Spanish readers was also ascribed to the disagreement regarding southern otoliths. The less clear ring pattern and presence of false rings is responsible for the poor ageing agreement of southern otoliths. False rings are more evident in the anti-rostrum of the otolith. This was the area recommended for age reading in the previous workshop. However, to overcome the problem of false rings it is advisable to use the rostrum as the reading area.

The birthdate criterium was not anticipated as a question to this workshop. However, it came up as an important issue during the discussions. As a consequence of the adoption of the 1<sup>st</sup> of January as the birthdate, zero age group is not assigned during the first semester of the year. Young fish frequently found off the Portuguese coast during the first semester are aged as one year olds even if their otoliths do not show any marked annulum. Fish from the same cohort caught during the second semester are assigned to the 0-group, leading to the separation of cohorts that are the same. This subject has already been dealt with by Azevedo (1999) and the workshop considers that its implications for sardine stock assessment should be discussed in more detail by ICES Working Group on the Assessment of Mackerel, Horse Mackerel, Sardine and Anchovy.

This workshop recommends that regular exchanges of otoliths should continue among the countries involved in the monitorisation of sardine stock. The preparation and regular circulation of an otolith reference collection covering the different areas and seasons, in general should contribute to improve age reading performance and to curtail the problem of within reader consistency. The protocol for sardine age readings was changed to accommodate the conclusions of the current workshop (Annex 2).

## ACKNOWLEDGEMENTS

We thank to all the people involved in the otolith exchange, those who participated in the age readings and also those who helped in the preparation of the otolith collections. This work was funded by Project PELASSES (EU DG FISH, Project no. 99/010).

## REFERENCES

- AZEVEDO, M., 1999. Exploratory data analysis for Iberian Sardine (*Sardina pilchardus*). Working Document to the Working Group on the Assessment of Mackerel, Horse Mackerel, Sardine and Anchovy., Copenhagen 14-23 September 1999. 13 p.
- ELTINK, A.T.G.W., 2000. Age reading comparisons. (MS Excel workbook version 1.0 October 2000) Internet: <http://www.efan.no>.
- ELTINK, A.T.G.W.; NEWTON, A.W.; MORGADO, C.; SANTAMARIA, M.T.G.; MODIN, J. 2000. Guidelines and tools for age reading comparisons. European Fish Ageing Network (EFAN) Report 3-2000, 73 p.
- ICES, 1997. Report of the Workshop on sardine otolith age reading. ICES CM 1997/H:7, 47p.

**ANNEX 1**  
Sardine age readings from the otolith exchange

Stratum	Sample no	Fish no	Fish length	Landing month	AJ Reader 1	IR Reader 2	PC Reader 3	ED Reader 4	QP Reader 5	EV Reader 6	Modal age	Percent agreement	Precision CV
IXaN	1	1	11,8	13-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	2	11,9	13-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	4	12,4	13-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	6	12,3	13-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	13	12,8	13-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	17	12,9	13-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	24	13,1	13-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	27	13,3	13-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	36	13,5	13-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	44	14,4	13-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	47	14,3	13-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	56	14,6	13-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	64	15,0	13-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	77	15,7	13-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	86	16,2	13-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	97	16,8	13-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	100	17,1	13-03-00	2	2	1	1	1	2	2	50%	37%
IXaN	1	1	11,4	14-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	33	13,9	14-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	79	16,8	14-03-00	2	2	1	2	1	2	2	67%	31%
IXaN	1	85	17,2	14-03-00	2	2	1	2	1	2	2	67%	31%
IXaN	1	88	18,3	14-03-00	2	2	2	2	2	2	2	100%	0%
IXaN	1	89	19,4	14-03-00	5	4	3	3	3	3	3	67%	24%
IXaN	1	121	17,7	14-03-00	2	2	2	1	2	2	2	83%	22%
IXaN	1	133	18,4	14-03-00	3	3	1	2	3	3	3	67%	33%
IXaN	1	141	18,6	14-03-00	3	2	2	3	2	2	2	67%	22%
IXaN	1	154	19,4	14-03-00	4	2	2	3	5	4	4	33%	36%
IXaN	1	158	19,6	14-03-00	3	2	2	3	3	2	3	50%	22%
IXaN	1	160	20,2	14-03-00	4	4	3	4	4	4	4	83%	11%
IXaN	1	25	14,7	15-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	35	15,1	15-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	44	15,6	15-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	56	16,4	15-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	1	84	17,8	15-03-00	2	2	1	2	2	2	2	83%	22%
IXaN	1	106	18,9	15-03-00	2	3	2	2	2	2	2	83%	19%
IXaN	1	124	19,7	15-03-00	3	4	2	3	3	3	3	67%	21%
IXaN	1	135	20,2	15-03-00	4	4	3	4	4	4	4	83%	11%
IXaN	1	144	20,7	15-03-00	3	3	3	4	3	4	3	67%	15%
IXaN	1	150	20,7	15-03-00	4	3	3	4	3	4	4	50%	16%
IXaN	1	157	21,0	15-03-00	7	7	6	6	6	6	6	67%	8%
IXaN	1	161	21,2	15-03-00	4	5	4	6	4	4	4	67%	19%
IXaN	1	167	21,5	15-03-00	3	4	3	6	4	5	3	33%	28%
IXaN	1	171	21,5	15-03-00	5	5	5	6	5	5	5	83%	8%
IXaN	1	175	22,3	15-03-00	6	6	5	7	6	6	6	67%	11%
IXaN	1	178	22,0	15-03-00	6	6	-	7	6	6	6	80%	7%
IXaN	2	1	11,4	21-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	2	3	11,3	21-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	2	12	11,8	21-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	2	14	11,8	21-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	2	21	12,0	21-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	2	22	12,0	21-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	2	31	12,5	21-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	2	32	12,5	21-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	2	41	13,0	21-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	2	42	13,1	21-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	2	51	13,9	21-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	2	52	13,6	21-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	2	54	14,6	21-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	2	1	14,0	21-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	2	4	14,3	21-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	2	6	14,9	21-03-00	1	2	1	1	1	1	1	83%	35%
IXaN	2	7	14,7	21-03-00	1	2	1	1	1	1	1	83%	35%
IXaN	2	16	15,0	21-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	2	18	15,4	21-03-00	1	2	1	1	1	1	1	83%	35%
IXaN	2	26	15,5	21-03-00	1	2	1	2	1	1	1	67%	39%

**ANNEX 1**  
Sardine age readings from the otolith exchange (continued)

Stratum	Sample no	Fish no	Fish length	Landing month	AJ Reader 1	IR Reader 2	PC Reader 3	ED Reader 4	QP Reader 5	EV Reader 6	Modal age	Percent agreement	Precision CV
IXaN	2	27	15,9	21-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	2	36	16,3	21-03-00	2	2	1	1	1	1	1	67%	39%
IXaN	2	37	16,0	21-03-00	2	2	1	2	1	1	2	50%	37%
IXaN	2	45	16,8	21-03-00	1	2	1	1	1	1	1	83%	35%
IXaN	2	47	16,8	21-03-00	1	2	1	1	1	1	1	83%	35%
IXaN	2	56	17,3	21-03-00	1	2	1	2	1	1	1	67%	39%
IXaN	2	57	17,1	21-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	2	66	17,6	21-03-00	2	2	1	1	1	1	1	67%	39%
IXaN	2	67	17,8	21-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	2	77	18,1	21-03-00	2	2	1	2	1	1	2	50%	37%
IXaN	2	79	18,3	21-03-00	2	2	2	2	2	2	2	100%	0%
IXaN	2	87	18,7	21-03-00	2	2	2	2	2	2	2	100%	0%
IXaN	2	89	18,9	21-03-00	2	2	2	2	3	2	2	83%	19%
IXaN	2	94	19,4	21-03-00	3	3	3	2	3	3	3	83%	14%
IXaN	2	95	19,4	21-03-00	5	4	5	3	4	4	4	50%	18%
IXaN	2	103	19,9	21-03-00	4	3	3	3	3	4	3	67%	15%
IXaN	2	105	19,7	21-03-00	5	4	4	4	5	4	4	67%	12%
IXaS	3	5	17,4	29-03-00	2	2	1	2	1	2	2	67%	31%
IXaS	3	12	17,9	29-03-00	3	3	3	2	2	2	3	50%	22%
IXaS	3	20	18,4	29-03-00	3	3	3	3	3	3	3	100%	0%
IXaS	3	29	18,8	29-03-00	3	3	2	3	2	2	3	50%	22%
IXaS	3	38	19,0	29-03-00	3	4	3	3	3	3	3	83%	13%
IXaS	3	47	19,9	29-03-00	8	9	5	5	9	5	5	50%	30%
IXaS	3	55	20,4	29-03-00	6	6	5	6	4	5	6	50%	15%
IXaS	3	57	20,4	29-03-00	8	7	6	6	9	8	8	33%	17%
IXaS	3	67	20,9	29-03-00	8	7	6	7	9	6	7	33%	16%
IXaS	3	69	20,7	29-03-00	7	6	4	6	7	5	7	33%	20%
IXaS	3	75	21,2	29-03-00	7	8	5	7	9	5	7	33%	23%
IXaS	3	79	21,2	29-03-00	6	6	5	6	6	6	6	83%	7%
IXaS	3	86	21,8	29-03-00	8	7	7	7	9	8	7	50%	11%
IXaS	3	88	21,5	29-03-00	8	8	8	8	7	8	8	67%	8%
IXaS	3	98	22,4	29-03-00	7	6	5	7	9	8	7	33%	20%
IXaS	3	103	22,3	29-03-00	8	9	8	8	9	9	8	50%	6%
IXaS	3	110	23,0	29-03-00	7	8	5	7	8	7	7	50%	16%
IXaS	3	112	23,2	29-03-00	7	8	6	8	8	7	8	50%	11%
IXaS	3	8	19,5	30-03-00	6	6	4	5	6	5	6	50%	15%
IXaS	3	46	21,7	30-03-00	8	8	5	8	11	7	8	50%	25%
IXaS	3	2	16,4	30-03-00	2	2	2	2	1	1	2	67%	31%
IXaS	3	4	16,0	30-03-00	2	2	1	2	1	1	2	50%	37%
IXaS	3	11	16,7	30-03-00	2	2	1	2	1	1	2	50%	37%
IXaS	3	21	17,0	30-03-00	1	2	1	2	1	1	1	67%	39%
IXaS	3	43	18,2	30-03-00	2	3	2	3	2	2	2	67%	22%
IXaS	3	53	18,8	30-03-00	2	2	2	3	2	2	2	83%	19%
IXaS	3	2	16,9	31-03-00	2	1	2	1	2	2	2	67%	31%
IXaS	3	21	17,8	31-03-00	3	2	3	2	3	2	3	50%	22%
IXaS	3	109	22,9	31-03-00	6	9	5	6	8	10	6	33%	27%
IXaS	3	110	22,5	31-03-00	6	7	5	6	9	8	6	33%	22%
IXaS	3	31	19,1	31-03-00	3	2	2	3	3	3	3	67%	19%
IXaS	4	101	21,4	06-03-00	4	6	3	8	3	4	4	33%	42%
IXaS	4	103	21,1	06-03-00	6	6	5	6	6	6	6	83%	7%
IXaS	4	111	21,7	06-03-00	5	5	4	7	4	5	5	50%	22%
IXaS	4	113	21,6	06-03-00	5	6	4	7	6	5	5	33%	19%
IXaS	4	1	14,4	06-03-00	1	1	2	1	2	1	1	67%	39%
IXaS	4	9	14,8	06-03-00	2	1	2	1	1	1	1	67%	39%
IXaS	4	11	14,6	06-03-00	2	1	1	1	1	1	1	83%	35%
IXaS	4	19	15,3	06-03-00	2	1	1	1	1	1	1	83%	35%
IXaS	4	21	15,3	06-03-00	2	1	1	1	1	1	1	83%	35%
IXaS	4	31	15,9	06-03-00	2	1	1	1	1	1	1	83%	35%
IXaS	4	33	15,9	06-03-00	2	2	2	1	1	1	2	50%	37%
IXaS	4	40	16,0	06-03-00	2	2	1	1	1	1	1	67%	39%
IXaS	4	41	16,2	06-03-00	1	1	1	1	1	1	1	100%	0%
IXaS	4	49	16,9	06-03-00	2	2	2	2	1	2	2	83%	22%
IXaS	4	51	16,5	06-03-00	2	2	1	2	2	1	2	67%	31%
IXaS	4	59	17,3	06-03-00	2	2	1	2	1	2	2	67%	31%
IXaS	4	61	17,2	06-03-00	2	1	1	1	2	1	1	67%	39%
IXaS	4	69	17,8	06-03-00	4	2	2	2	3	2	2	67%	33%
IXaS	4	70	17,6	06-03-00	3	1	2	1	3	1	1	50%	54%



**ANNEX 1**  
Sardine age readings from the otolith exchange (continued)

Stratum	Sample no	Fish no	Fish length	Landing month	AJ Reader 1	IR Reader 2	PC Reader 3	ED Reader 4	QP Reader 5	EV Reader 6	Modal age	Percent agreement	Precision CV
VII	6	9	26,4	21-03-00	6	6	7	8	9	11	6	33%	25%
VII	6	10	17,6	21-03-00	1	1	1	2	1	1	1	83%	35%
VII	6	11	18,3	21-03-00	1	1	1	1	1	1	1	100%	0%
VII	6	12	24,7	21-03-00	7	8	6	8	11	10	8	33%	22%
VII	6	13	15,8	21-03-00	1	1	1	1	1	1	1	100%	0%
VII	6	14	19,6	21-03-00	2	2	2	2	2	2	2	100%	0%
VII	6	15	16,6	21-03-00	1	1	1	1	1	1	1	100%	0%
VII	6	16	24,4	21-03-00	8	8	8	8	11	10	8	67%	15%
VII	6	17	25,0	21-03-00	6	5	6	8	10	9	6	33%	27%
VII	6	18	22,9	21-03-00	3	3	3	4	3	4	3	67%	15%
VII	6	19	24,0	21-03-00	7	7	7	7	7	7	7	100%	0%
VII	6	20	15,8	21-03-00	1	1	1	1	1	1	1	100%	0%
VII	6	21	23,5	21-03-00	3	3	3	3	4	3	3	83%	13%
VII	6	22	21,5	21-03-00	2	2	2	3	3	2	2	67%	22%
VII	6	23	19,2	21-03-00	3	2	2	3	3	2	3	50%	22%
VII	6	24	23,5	21-03-00	3	3	3	3	3	3	3	100%	0%
VII	6	25	24,7	21-03-00	4	4	5	4	5	5	4	50%	12%
VII	6	26	17,0	21-03-00	1	1	1	1	2	1	1	83%	35%
VII	6	27	22,3	21-03-00	-	4	4	4	4	5	4	80%	11%
VII	6	28	15,4	21-03-00	1	1	1	1	1	1	1	100%	0%
VII	6	29	15,0	21-03-00	1	1	1	1	1	1	1	100%	0%
VII	6	30	17,0	21-03-00	1	1	1	1	2	1	1	83%	35%
IXaN	7	1	17,1	29-03-00	1	2	1	1	3	1	1	67%	56%
IXaN	7	2	16,5	29-03-00	2	2	1	2	1	2	2	67%	31%
IXaN	7	3	15,6	29-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	7	4	16,4	29-03-00	2	2	1	2	1	2	2	67%	31%
IXaN	7	5	15,4	29-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	7	6	16,0	29-03-00	2	2	1	2	1	2	2	67%	31%
IXaN	7	7	18,5	29-03-00	2	1	1	2	3	1	1	50%	49%
IXaN	7	8	15,5	29-03-00	1	2	1	1	1	1	1	83%	35%
IXaN	7	9	17,6	29-03-00	2	3	2	2	3	3	2	50%	22%
IXaN	7	10	17,2	29-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	7	11	16,9	29-03-00	2	2	1	1	1	2	2	50%	37%
IXaN	7	12	19,4	29-03-00	3	3	3	3	2	2	3	67%	19%
IXaN	7	13	17,5	29-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	7	14	15,1	29-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	7	15	18,2	29-03-00	4	3	2	2	2	2	2	67%	33%
IXaN	7	16	19,6	29-03-00	4	4	3	3	3	3	3	67%	15%
IXaN	7	17	19,1	29-03-00	2	2	2	2	2	2	2	100%	0%
IXaN	7	18	19,5	29-03-00	3	4	3	3	3	4	3	67%	15%
IXaN	7	19	18,5	30-03-00	2	1	1	2	1	1	1	67%	39%
IXaN	7	20	21,0	30-03-00	4	3	3	4	3	3	3	67%	15%
IXaN	7	21	18,1	30-03-00	1	1	1	1	1	1	1	100%	0%
IXaN	7	22	21,0	30-03-00	3	3	2	3	4	4	3	50%	24%
IXaN	7	23	20,0	30-03-00	2	2	2	2	3	2	2	83%	19%
VIII	8	1	23,5	31-03-00	5	5	6	6	9	8	5	33%	25%
VIII	8	2	22,6	31-03-00	3	4	4	4	4	4	4	83%	11%
VIII	8	3	22,7	31-03-00	4	3	4	4	4	5	4	67%	16%
VIII	8	4	21,3	31-03-00	3	2	2	3	2	2	2	67%	22%
VIII	8	5	22,2	31-03-00	4	4	4	3	4	4	4	83%	11%
VIII	8	6	21,3	31-03-00	3	2	2	3	3	3	3	67%	19%
VIII	8	7	23,5	31-03-00	6	4	6	4	7	7	6	33%	24%
VIII	8	8	21,5	03-04-00	4	4	4	3	4	4	4	83%	11%
VIII	8	9	23,0	03-04-00	6	6	6	6	6	8	6	83%	13%
VIII	8	10	22,4	03-04-00	4	4	5	3	5	5	5	50%	19%
VIII	8	11	20,9	03-04-00	4	4	4	3	4	5	4	67%	16%
VIII	8	12	23,0	03-04-00	4	4	4	4	4	4	4	100%	0%
VIII	8	13	21,9	03-04-00	3	3	3	3	3	3	3	100%	0%
VIII	8	14	20,8	03-04-00	4	4	3	3	4	4	4	67%	14%
VIII	9	1	20,7	26-04-00	3	3	3	3	3	3	3	100%	0%
VIII	9	2	20,8	26-04-00	4	4	4	3	4	4	4	83%	11%
VIII	9	3	23,0	26-04-00	7	8	7	6	8	8	8	50%	11%
VIII	9	4	22,2	26-04-00	6	5	6	6	7	7	6	50%	12%
VIII	9	5	23,2	26-04-00	6	6	6	6	6	7	6	83%	7%
VIII	9	6	23,6	06-04-00	6	6	7	5	7	7	7	50%	13%
VIII	9	7	25,4	06-04-00	5	5	5	6	8	7	5	50%	21%
VIII	9	8	24,1	06-04-00	6	4	6	6	11	7	6	50%	35%
VIII	9	9	20,3	08-04-00	2	2	2	4	3	2	2	67%	33%



**ANNEX 1**  
**Sardine age readings from the otolith exchange (continued)**

Stratum	Sample no	Fish no	Fish length	Landing month	AJ Reader 1	IR Reader 2	PC Reader 3	ED Reader 4	QP Reader 5	EV Reader 6	Modal age	Percent agreement	Precision CV
VIII	12	4	20,3	13-04-00	2	2	2	2	2	2	2	100%	0%
VIII	12	5	19,2	13-04-00	2	2	2	2	3	2	2	83%	19%
VIII	12	6	19,0	13-04-00	2	2	2	2	3	2	2	83%	19%
VIII	12	7	21,6	13-04-00	3	3	2	3	4	5	3	50%	31%
VIII	12	8	18,5	13-04-00	2	2	2	2	2	2	2	100%	0%
VIII	12	9	22,6	13-04-00	8	8	8	7	10	10	8	50%	14%
VIII	12	10	18,0	13-04-00	2	2	2	2	2	2	2	100%	0%
VIII	12	11	18,5	13-04-00	2	2	2	2	2	2	2	100%	0%
VIII	12	12	20,5	13-04-00	3	3	3	3	3	4	3	83%	13%
VIII	12	13	20,8	13-04-00	2	2	2	2	2	2	2	100%	0%
VIII	12	14	21,3	13-04-00	3	3	3	3	3	3	3	100%	0%
VIII	12	15	21,0	13-04-00	4	2	2	2	3	2	2	67%	33%
VIII	12	16	20,3	13-04-00	4	6	7	7	10	9	7	33%	30%
VIII	12	17	23,6	13-04-00	5	6	6	4	6	7	6	50%	18%
VIII	12	18	21,5	13-04-00	-	-	4	3	4	4	4	75%	13%
VIII	12	19	22,5	13-04-00	5	5	5	4	6	6	5	50%	15%
VIII	12	20	23,0	13-04-00	5	5	5	4	5	5	5	83%	8%
VIII	12	21	22,0	13-04-00	3	3	3	3	5	5	3	67%	28%
VIII	12	22	23,0	13-04-00	6	6	6	6	6	7	6	83%	7%
Total read					354	356	357	357	357	357		76,1%	15,5%
Total NOT read					3	1	0	0	0	0			

**ANNEX 2****PROTOCOL FOR SARDINE AGE DETERMINATION**

In order to standardise the sardine age assignments and to improve the age estimates the Workshop held in Lisbon-Portugal, 2002 adopted the following protocol:

1. The first of January is adopted as the birthdate reference for age assignment purposes. Consequently, if an otolith is collected from a fish caught in the first semester of the year the age group assignment will correspond to the number of hyaline zones present. If the otolith is extracted from a fish caught in the second semester of the year the age group assigned will correspond to the hyaline zones completely formed, i.e. if the edge of the otolith is hyaline it will be not considered.
2. After extraction otoliths are washed thoroughly, dried, mounted and preserved in xylol resistant plastic plaques in a synthetic resin (“Eukitt” or “Entellan”).
3. The observations of entire otoliths are made under reflected light against a black background using magnifying dissection microscopes with 20X magnification. Magnification should be increased near the otolith edge to improve the discrimination of narrow hyaline rings in older individuals.
4. It is always advisable to have pairs of whole otoliths available from individual sardine specimens when trying to interpret the ring structure.
5. A set of an opaque and a hyaline zone corresponds to one annual growth zone (*annulus*).
6. It is recommended to use the *anti-rostrum* as the most adequate zone to count hyaline rings for age assignment. However, since false rings are more evident in this region, the *rostrum* should be used in otoliths from the southern area.
7. Sometimes it may happen that other areas of the otolith, i.e. the dorsal part, are easier to read. In this case the age reading based on the analysis of these areas can be considered appropriate if the readings prove to be consistent.
8. In order to adopt a ring as an *annulus* it is recommended that the ring can be followed throughout the whole otolith contour. This rule must be applied specially for the first three *annuli*, since in the older specimens growth often slows down to such an extent that hyaline rings are very close together. In that case opaque and hyaline zones become more difficult to be identified.