

WORKSHOP ON AGE READING OF BLUE WHITING (WKARBLUE3; OUTPUTS FROM 2021 MEETING)

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WORKSHOP ON AGE READING OF BLUE WHITING (WKARBLUE3; OUTPUTS FROM 2021 MEETING)

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i Executive summary

The workshop on age reading of Blue whiting (WKARBLUE3) was conducted to go through the following objectives: review, document and make recommendations on current methods of aging blue whiting (*Micromesistius poutassou*) to produce the input data for this species stock assessment.

The workshop was preceded by an otolith exchange on SmartDots (Event ID 278). The exchange included 407 otolith images, 190 from the northern areas (ICES divisions 2.a, 4.a, 5.a, 5.b, 6, 6.a, 6.b, 7 and 7.c) and 217 from the southern areas (ICES divisions 9.a and 8.c). A total of 27 readers from 11 countries participate in the exchange, 8 where from the southern areas and 19 from the northern areas. The overall agreement with the modal age from the pre-workshop exercise was 66%. Although, considering only the results from the northern areas by the advanced readers that usually read the otoliths from those areas for the assessment, the percentage of agreement was 69%. In relation to the otoliths from the southern areas, the percentage of agreement on age classifications by the advanced readers that read for the assessment was 79%.

The pre-workshop exchange results reveal that readers are prone to be off in interpreting the age of fish in other areas which they don't normally read for the stock assessment. The reason to this discrepancy is that blue whiting presents a wide distribution along the Northeast Atlantic with growth variations between areas. The blue whiting from the northern areas of the stock (North of the British Isles) has a slower growth than in the more southern areas (south of the British Isles). The otoliths from some of the northern areas (2.a, 5.b, 6.b and 7.c) were more difficult to read. These results were also observed in the data analysis when only the readers that usually read the otoliths from the northern areas were included.

During the workshop a new small exchange was also conducted, which included 22 otoliths from the southern areas and 33 from the northern areas (SmartDots EventID 347). In this new exchange the readers only read the otoliths from the areas they usually read from in the assessment. The results showed a slightly increase in the percentage of agreement, with a 78% of agreement in the otoliths from the southern areas and a 71% in the otoliths from the northern areas.

The other main issues identified during this workshop were the position of the first annual growth ring, false or split rings and interpretation of the edge. These issues are the same as has been mentioned in previous reports and workshops, and thus a reoccurring problem among age readers. A reference collection of images with annotations will be uploaded to SmartDots as soon as the working group on SmartDots governance (WGSMART) has the reference collection module ready.

The realization of age reading workshops on blue whiting constitute an advantage and are recommended to be conducted on a regular basis, to increase the precision on aging between readers. Nevertheless, since the main issues on aging blue whiting still remain, validation studies applied to this species are also recommended and needed to allow the increasing of accuracy on age classifications.

ii Expert group information

Expert group name	Workshop on Age estimation of Blue Whiting (<i>Micromesistius poutassou</i>) (WKARBLUE3)
Expert group cycle	N/A
Year cycle started	N/A
Reporting year in cycle	1/1
Chair(s)	Patrícia Gonçalves, Portugal
	Jane A. Godiksen, Norway
Meeting venue(s) and dates	31 st May – 4 th June 2021, Online (23 participants)

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1 Introduction

Blue whiting (*Micromesistius poutassou*) is a pelagic gadoid which is widely distributed in the eastern part of the North Atlantic. The highest concentrations are found during spawning along the edge of the continental shelf in areas west of the British Isles and on the Rockall Bank plateau where it occurs in large schools at depths ranging between 300 and 600 meters but is also present in almost all other management areas between the Barents Sea and the Strait of Gibraltar and west to the Irminger Sea. Adults reach maturation at 1-3 years old (ICES 2013a) and undertake long annual migrations from the feeding grounds to the spawning grounds (Bailey, 1982). Most of the spawning takes place between March and April, along the shelf edge and the banks west of the British Isles. Juveniles are abundant in many areas, with an important nursery area believed to be the Norwegian Sea, at least in times of high abundance. Morphological, physiological, and genetic research has suggested that there may be several components of the stock which mix in the spawning area west of the British Isles. Due to the large population size, its considerable migratory capabilities and wide spatial distribution, the stock composition and dynamics require continued monitoring.

Prior to 1993, for the purposes of assessment, it was assumed that blue whiting had two components, a northern and a southern component. The Northern stock was known to feed in the Norwegian Sea and spawn to the west of the British Isles. The Southern stock was found along the continental shelf off the coast of Spain and Portugal with the main spawning areas towards the Porcupine Bank. The Porcupine Bank was considered a transitional area between the two main stocks (ICES, 1990). In 1993 it was argued that there was no strong evidence to maintain this division between the two stocks. Results from an otolith age reading workshop at that time showed no significant difference in mean annual ring diameter between northern and southern stocks. It was agreed by ACFM in 1993 that the two stocks should be combined for assessment purposes (ICES, 1995). Since then this stock has been assessed as one unit.

Several approaches have been employed to investigate the stock structure of blue whiting. The studies relating to genetics revealed genetic variability between the populations from the Northeast Atlantic and the Mediterranean (Mork and Giæver, 1995; Giæver and Stein, 1998; Ryan *et al* 2005) and from the Celtic Sea and the Bay of Biscay (Was *et al*. 2008). Studies on larval otolith growth patterns (Brophy and King, 2007) revealed differences in growth between Northern spawning areas and the Bay of Biscay. Several studies on the movements of eggs and larvae, and on otoliths shape analysis (Bartsch and Coombs, 1997; Skogenet *et al.*, 1999; Pointin and Payne, 2014; Mahe *et al.*, 2016; Keating *et al.*, 2014) gave results which showed that the southern stock will spawn in an area where the eggs and larvae are likely to drift southwards and the northern stock where the eggs and larvae will drift northwards. Spawning starts earlier in the southern area (by at least one month), with peak spawning occurring later further north and also the existence of two distinct morphology types, from fish occupying distinct geographical distribution areas (northern and southern areas). The most recent studies support the hypothesis of northern and southern area blue whiting population which may overlap to varying degrees in the centre of the spawning distribution.

Taking into account those numerous scientific studies suggesting that blue whiting in the North Atlantic consists of multiple stock units, the ICES Stock Identification Methods Working Group (SIMWG) reviewed this evidence in 2014 (ICES, 2014) and concluded that the perception of blue whiting in the NE Atlantic as a single-stock unit is not supported by the best available science. SIMWG further recommended that blue whiting be considered as two units. However, there is currently no information available that can be used as the basis for generating advice on the status of the individual stocks.

Although, for the assessment purposes the stock is still considered a single unit across the Northeast Atlantic, differences in growth rate and maturation have implications on otolith growth increments deposition between the samples from the southern and the northern areas. Those differences had been taken into account during the data analysis of the age classifications from the pre-workshop exchange (SmartDots EventID 278) and the results are also presented separately by areas.

2 Review information on age estimations and validation work (ToR a and b)

2.1 Validation/corroboration studies on Blue whiting

Blue whiting a widely distributed species, shows a slower growth pattern when moving north along the Northeast Atlantic (NEA) (e.g. Gonçalves et al. 2017). The peak of spawning is also known to be different between the southern (January-March) and the northern areas (March-April). The main age reading guidelines for this species are common for all the NEA. One of the most important guideline, is the one that describes the size of first ring which should be consider around 50 - 56 e.p.u. (8.33 – 9.33 mm) to avoid counting the Baileys zone (larval ring zone). Besides, the main source of uncertainties on age reading for this species result from the common age reading interpretation for the whole NEA distribution area, like the first ring interpretation, due to misinterpretation with the Bailey's zone and also the difficult on false and double rings identification. Taking this into account age corroboration studies had been prepared and presented during the 2017 workshop on blue whiting age reading (ICES WKARBLUE 2) with the intent to help on the clarification on the age rings interpretation (see details in ICES 2017). Those studies had been mainly focused on length based methods and back calculation analysis:

(a) for the <u>identification of the first ring</u> on otoliths collected off the Portuguese coast (ICES division 27.9.a) (Dores and Gonçalves, 2017) (see also section 4.1 in ICES 2017) (Section 2.1.1);

(b) based on a multivariate modelling approach that have been applied to help on the <u>identifica-</u> <u>tion of false and split rings</u>, this resulted in the so called "IPMA table" (Gonçalves and Dores, 2017) (see also section 4.2 in ICES 2017) (Section 2.1.2);

(c) <u>marginal increment analysis</u> (Elleboode and Chantre, 2017) (see also section 4.3 in ICES 2017) (Section 2.1.3).

2.1.1 Portuguese Coast 1st ring length validation

A study on the first ring validation was conducted by IPMA applied to the blue whiting age reading from the Portuguese coast (Dores and Gonçalves, 2017) (see details in section 4.1 in ICES 2017). For this study, a sample of 67 otoliths was selected. From those otoliths age was determined, and the following measurements recorded: otolith length (mm), otolith width (mm), otolith thickness (mm), age ring diameter (mm) and age ring width (mm). One otolith from each pair used in age reading was burned. Digitized images of both otoliths were obtained. Furthermore, all the 67 otolith pairs (1 burned and 1 not burned) were also sliced and digitized images were obtained.

The results showed a higher correlation of the modal age with otolith thickness with otolith length (Figure 2.1).

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[1]



Figure 2.1. Relationships obtained between fish length, otolith length, otolith width, otolith thickness and modal age. The numbers represent the R-value from the adjusted model between the variables.

The relationship between otolith total length (Y) and fish length (x) was established in the present work, by the following equation [1]:

Y=2.796(x)-7.219 (R²=0.99)

Equation [1] allows the estimation of the total fish length at the time of formation of each annual ring. Figure 2.2 shows for each otolith the total length of each age ring and the estimated mean fish length for all the age rings determined through the equation [1].



Figure 2.2 (a) Otolith ring length (mm) and (b) fish length (cm) at each age group from 1 - 6. In green the estimated measurements for the first ring length (a) and for the fish length at age 1 (b).

The size of first ring on otoliths off the Portuguese coast (southern area) varies between 8.5-11 mm in diameter (Figure 2.2), those lengths are different from the ones described on literature based on blue whiting measurements from the northern areas (8.3-9.3 mm), which corresponds to an estimated fish total length between 17.9 and 20.7 cm.

In order to test the different preparation techniques, one of the otoliths from each pair was burned to clarify growth rings and improve age reading. As a result, in some otoliths the two inner growth rings previously marked as distinct annual rings by age readers seemed to fuse, apparently forming only one band (Figure 2.3).



Figure 2.3 - Otoliths pair from a 30.7 cm total length fish. The otolith on top is not burned and has the two first annual rings length measurements. The otolith below shows that the first two rings apparently have been fused during the burning process.

After burning, it is not possible to visualize the internal structure near the nucleus of larger otoliths, as the two inner zones seems to have been fused. This is probably the reason why the first annual ring often is measured larger as the fish increases in age. The question for the readers is whether the inner ring – which are not seen on the burned otolith – should be counted as a true age ring or not. T

The sliced otoliths have been also analysed but this technique doesn't seem to improve the age readings on this species. Taking into account that the process of preparing and make the slicing of the otoliths is time consuming, no benefits on this application have been identified.

2.1.2 Multivariate modelling approach to help on the identification of false and split rings

The modal age from the results of the 2017 pre-workshop calibration exercise (ICES 2017) were used to define more objective criteria to help readers to improve the accuracy and precision of the blue whiting age reading classifications. For each of the 245 otoliths pairs, otolith total length, otolith weight and age rings diameters were determined. Also, the data from the fish total length and the ICES area where fish were caught was used.

The results showed that the relationship between the modal age with fish length, otolith length and otolith weight was linear (Figure 2.4).



Figure 2.4. Scatterplot of the relationship between all the variables: fish total length (line 1, column 1); 1st ring diameter (line 2, column 2); otolith length (line 3, column 3); otolith weight (line 4, column 4); and modal age (line 5, column 5).

The relation between the 1st ring diameter and all the other variables presents a linear trend, with the exception with the modal age in which no clear pattern was evident. The 1st ring diameter shows an increase with the increase in fish total length, otolith length and otolith weight. This increase of 1st ring diameter when compared with the other variables gives indication of the existence of some problems with the interpretation and the identification of the inner ring across

the ages. These results go in line with the observations made when applying the burning techniques and described in Section 2.1.1.

The modal age by fish length and by area was analysed, but the results reveal that there could be some problems in the age classification by area. In some of those ICES areas the age-length curve does not present a shape similar to the von Bertalanffy growth curve model, mainly in 8.c, 7.c, 6.a, 2.b and 14.b. This could be due to difficulties in age classification of the otoliths from those areas or due to a small sample size, or a mix of the two reasons (see section 4.2 in ICES 2017 for details).

Taking into account the growth pattern of blue whiting described for the Northeast Atlantic (Bailey, 1982; ICES, 2016) and the relationship between the age, the fish total length and otolith length an approach based on multivariate modelling has been applied to validate ages and identify false and/or split rings (Figure 2.5).



Figure 2.5. Relation between the fish length (mm) and the otolith length (mm) by modal age (in colours) from (a) WKARBLUE2 and (b) estimated by a multivariate modelling approach. Fish ID is identified in numbers. Identified with a red square the Fish ID 74 classified as age 4 (modal age), but according with the multivariate model it should be classified as an age 3, which indicates the existence of a double or split/false ring.

This approach has showed to be useful to help on age rings interpretation, mainly concerning the doubts due to false and double rings and resulted in the so called IPMA table. The first version of the "IPMA table", was proposed with the aim to provide length measurements estimations on the different age rings, from ages 0 to 10. The main conclusion during the 2017 workshop was that the IPMA criteria table seems to work, but the limited sample size (by ICES area and sex), do not allowed to obtain the length measurements estimations by area and sex with the suitable precision. A new criteria table based on more data samples (by area, quarters and sexes) should be proposed and tested during the next blue whiting age reading workshop (WKARBLUE3) (ICES, 2017).

The updated version of the IPMA table has been elaborated and is now presented in Section 2.2.

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2.1.3 Marginal increment analysis (MIA)

The results on the marginal increment analysis (MIA) applied to the 2017 blue whiting otoliths from the exchange had been inconclusive due to also a small sample size (ICES, 2017). Although, the MIA technique have proven to be helpful and accurate concerning the corroboration and validation of ageing and have been successful applied in several other species.

2.2 Validation study (IPMA) (Gonçalves and Silva, 2021)

On blue whiting the interpretation of age rings revealed to be a difficult task and the perception and interpretation of false/split rings could change among readers. The classification criteria's and guidelines are made to be objective and to help readers on their task, but the final age is always dependent on the experience of the reader and its interpretation of the otolith growth pattern. The regular calibration workshops aiming to improve precision and accuracy among readers. The expectation when conducting a intercalibration workshop is to obtain a percentage of agreement around 100%, but most of the times and special for the older ages the percentage is below 50%. Those results occur due to the difficulty into define objective criteria and to the fact that age interpretation could be compromised by the definition of growth rings, the spawning period, the feeding period or some physiological changes that could occur and have implications/changes on fish's growth. Although, all of those constraints on age pattern interpretation, the definition of objective criteria's may help readers to improve the agreement among them, mainly by identifying false and/or split rings. False or split rings have also been an issue on blue whiting aging along the years and have been reported in almost all age workshops.

To help readers on their task, mainly on false and/or split rings identification, the data from the 2020 pre-workshop exchange was analysed, taking into account the following variables: otolith length, fish total length, otolith weight, ICES area and the agreed modal age. A model was applied to the data, following the same approach has described in ICES 2017 (Gonçalves and Dores, 2017) (Section 4.2 in ICES 2017), with the addition/modification that the ages for each fish were estimated based on the application of a machine learning model. The estimated age was analysed by sex and by two main groups of ICES areas (northern and southern areas).

For this validation study the data from the otoliths collected under ICES areas from the 2020 preworkshop exchange were analysed (Figure 2.6).



Figure 2.6. Number of otolith samples by ICES area and by sex (orange – no sex information; F- females; I – Undetermined; M – males) from the pre-workshop exchange.

2.2.1 Results

The results from the data modelling were based on the comparison with age structure of the samples from the 2020 pre-workshop exchange using the modal age based on the readers' classifications *versus* the ages estimated by the application of the machine learning model evaluation algorithm (MLE) applied in the current validation study from IPMA (Figure 2.7 and 2.8).

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Results from the BW pre-wk Exchange 2020



Figure 2.7. Principal components analysis of the different age groups (0-8) projected in represented in two axes (PC1 and PC2) for the data from the pre-workshop exchange *versus* the ages estimated based on the model applied in the validation study from IPMA (MLE).



Figure 2.8. Relation between the fish length class (in millimeters) and the otoliths total length (in millimeters) by each age group (0-8) and by ICES area component (Northern and Southern areas) for the data from the pre-workshop exchange *versus* the ages estimated based on the model applied in the validation study from IPMA (MLE). The blue square identifies the modal ages classification for ages 0 to 3 years old.

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2.2.2 Conclusions

The readers have applied the IPMA Table that resulted from the MLE application to the northern and southern areas otoliths. The readers have mentioned that in some cases the ages observed do not correspond with the age estimated and proposed by the method.

The MLE has been trained with data from the 2020 pre-workshop exchange, mainly the modal ages. However, due to the high uncertainty on age classifications reflected in a lower percentage of agreement, this may have resulted in "low" quality data to feed and train the model.

The IPMA table for the Northern and Southern areas resulted from the MLE application is available in Annex 5.

Although, the main conclusion was not to apply it for now, due the fact that some uncertainties still remain, mainly because the age agreement on the otoliths from the exchange was only achieved on a reduced number of otoliths.

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3 Age reading exchange prior to WKARBLUE3

An exchange was initiated in 2020 to establish and evaluate the agreement on blue whiting otoliths aging between readers. The results were discussed during the first days of the workshop. The exchange was conducted using images in SmartDots and consisted of 190 images from the northern areas, 217 images from the southern areas and 90 images from the Mediterranean Sea.

Samples from Mediterranean were also included on the exchange, despite this area is outside the limits defined for the stock. The reason for including those samples was for readers from the Mediterranean be able to calibrate their classifications and get some feedback on growth interpretation from the blue whiting age readings experts that currently read the otoliths for the assessment. In the end, readers from the Mediterranean only participate on the pre-workshop exchange and couldn't join the workshop. Most of readers from the Atlantic areas have opted to not read the otoliths from the Mediterranean, due to their lack of experience on growth interpretation on those otoliths. Thus, due to the before mentioned reasons and also to the absence of participants from the Mediterranean on the workshop, the main discussions during the workshop were focused on otoliths collected at the Northeast Atlantic waters.

Important highlights from the results of the exchange were:

- a) The clear difference in growth patterns in otoliths from fish south of the British Isles compared to otoliths from fish in the northern part of the Northeast Atlantic (ICES areas above the Porcupine Bank), results in lower age agreement when readers read otoliths from areas they are not familiar with.
- b) Readers normally reading otoliths from the southern areas typically underestimate the age of the northern areas, while readers normally reading otoliths from the northern component typically overestimate the age of the southern areas. This clearly lower the percent agreement of the stock from an exchange (Section 3.1). This could be less problematic in an assessment situation, as readers would only read the otoliths from their own areas (Section 3.3 and 3.4).
- c) A recommendation from WKARBLUE3 is that readers will mainly read otoliths from their own areas in a future exchange, as this will give a better view on the issues in age reading of the stock.
- d) The inner ring determination is still one of the main issues on age classification on blue whiting. A validation study based on daily increments should be done previous to the next workshop.
- e) During the workshop the participants agreed to gather some measurements of total length otoliths from age 0 (Quarters 3 and 4) and age 1 (Quarters 1 and 2). The main goal is to help readers, on the inner ring interpretation, by having a table with some measurements which will be also included on the age reading guidelines (Annex 6).

3.1 Results Pre-workshop exercise

In the pre-workshop exchange, a total of 407 otolith samples were used, with 190 from the northern areas (ICES divisions 2.a, 4.a, 5.a, 5.b, 6, 6.a, 6.b, 7 and 7.c) and 217 from the southern areas (ICES divisions 9.a and 8.c). A total of 27 readers from 11 countries participate in the exchange, from those 8 where from the southern areas with only 4 advanced readers (reading for stock assessment) and 19 from the northern areas of which 13 were advanced readers. The results from the pre-workshop age calibration exercise displayed clear issues in perception of otolith

structure. The overall agreement was only 66% with a precision of 26% CV and an age percentage error (APE) of 17% (see details in Section 5.1.2). Although, if we consider only the results from the northern areas by the advanced readers that usually read the otoliths from those areas for the assessment, the percentage of agreement was 69%, with 11% APE (see details in Section 3.3). In relation to the otoliths from the southern areas, the percentage of agreement on age classifications by the advanced readers that usually read those otoliths for the assessment was 79%, with 14% APE (see details in Section 3.4).

The otoliths from some of the northern areas resulted more difficult to read and to interpret the growth rings, those areas are 2.a, 5.b, 6.b and 7.c. Those areas are also the areas were most of the catch-at-age data used for the assessment comes from, which need a special attention and a further investigation on growth interpretation, due to the impacts of possible errors may have in this species stock assessment.

The other main issues identified during this workshop were the position of the first annual growth ring, false or split rings and interpretation of the edge.

3.2 All the areas pre-workshop results

Summary of statistics: Total number of samples (NSample), coefficient of variance (CV), percentage of agreement (PA) and average percentage error (APE) for all ages and readers (All and advanced).

NSample	сv	РА	APE	Readers
407	26%	66%	17%	All
407	19%	69%	11%	Advanced readers

Considering all the areas, for all age readers combined, the relative bias was found to be minimal (-0.13), but for individual age-readers the relative bias varied from -1.45 to +0.42 (Table 3.1, Table 3.2, and Figure 3.1). This shows a significant over- and under-ageing of otoliths by age readers, and high bias was found both among experienced readers and trainees. For fish older than five there is a tendency of underestimating the ages, while overestimation seems to be the main problem among younger individuals. This is also evident on an individual basis where older fish were generally under-aged compared to modal age, while there was variation in whether a reader under- or overestimated the ages of younger fish. The under/over-ageing signifies systematic miss-interpretation of growth structures within the otolith. There is also clear bias among readers compared to the modal age. The precision (CV), percentage of agreement and the standard deviation (STDEV) by modal age for all readers (Figure 3.1) reveals that the agreement decreases below 50% from the age 6 onwards.

Table 3.1. All areas and all the readers. The relative bias (as the difference between the mean and modal age) per modal age and reader is presented, as well as the weighted mean relative bias per reader and the relative bias per modal age for all readers combined.

Modal age	R01 ES R02 IE R03 DI		R03 DK	R04 PT	R05 NL	R06 PT	R07 RU R08 RU		
0	0.06	0.07	0.07	0.00	0.52	0.26	0.23	0.03	
1	0.14	0.09	0.00	0.11	0.51	0.00	0.12	0.02	

Modal age	R01 ES	R02 IE	R03 DK	R04 PT	R05 NL	R06 PT	R07 RU	R08 RU	
2	0.24	0.36	-0.09	0.08	0.38	-0.16	0.06	-0.01	
3	0.01	0.16	-0.38	-0.03	0.15	-0.29	0.01	-0.02	
4	-0.52	0.61	-0.91	-0.26	0.31	-0.93	-0.17	-0.20	
5	-0.82	0.41	-1.51	-0.67	0.32	-1.62	-0.38	-0.36	
6	-1.54	0.54	-1.82	-0.85	0.44	-2.04	-0.54	-0.33	
7	-1.36	0.64	-1.82	-1.18	0.45	-2.27	-0.09	0.00	
8	-1.90	0.10	-2.11	-0.90	0.60	-2.20	-0.30	0.10	
9	-1.25	1.25	-0.88	-0.75	1.00	-2.00	-0.25	0.12	
10	-2.00	2.00	-1.00	-0.50	0.75	-3.50	-0.25	-0.25	
11	-1.00	2.00	-1.00	-1.00	0.00	-3.00	0.00	1.00	
12	-3.75	0.25	-2.75	-1.50	-0.50	-2.33	-1.25	-1.25	
13	-3.00	0.00	-0.50	-1.50	-1.00	-4.00	-2.00	0.50	
14	-	-	-	-	-	-	-	-	
15	-	-	-	-	-	-	-	-	
16	-	-	-	-	-	-	-	-	
17	-	-	-	-	-	-	-	-	
18	-	-	-	-	-	-	-	-	
19	-11.00	0.00	-3.00	-10.00	-1.00	-6.00	-5.00	0.00	
Weighted Mean	-0.39	0.33	-0.64	-0.25	0.35	-0.73	-0.10	-0.09	
Modal age	R09 FO	R10 NO	R11 NO	R12 FO	R13 DE	R14 ES	R15 NO	R16 DE	
0	0.19	0.00	0.19	0.33	0.10	0.00	0.03	0.39	
1	0.25	0.04	0.11	0.38	0.16	0.00	0.07	0.25	
2	0.27	0.05	0.03	0.54	0.42	-0.26	0.10	0.34	
3	0.30	0.12	0.11	0.26	0.14	-0.44	0.07	0.08	
4	0.43	0.22	0.09	0.44	0.02	-1.02	0.13	-0.07	
5	0.72	0.19	0.15	0.34	0.08	-0.76	0.10	-0.10	
6	0.41	-0.07	-0.15	0.50	0.07	-1.50	-0.11	-0.19	
7	1.45	0.55	0.09	1.00	0.18	-1.43	0.64	0.00	
8	0.30	0.90	-0.60	1.00	-0.10	-1.88	-0.10	-0.30	

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Modal age	R09 FO	R10 NO	R11 NO	R12 FO	R13 DE	R14 ES	R15 NO	R16 DE
9	1.88	0.57	0.00	1.12	-0.12	-1.40	0.00	-0.38
10	-0.75	1.00	0.50	-0.25	-0.50	-2.33	0.25	-0.75
11	0.00	0.00	0.00	0.00	-1.00	-3.00	0.00	0.00
12	0.00	0.25	-0.25	0.00	-1.25	-3.33	-0.50	-1.50
13	0.50	0.50	-0.50	0.50	-2.00	-2.00	-0.50	-2.00
14	-	-	-	-	-	-	-	-
15	-	-	-	-	-	-	-	-
16	-	-	-	-	-	-	-	-
17	-	-	-	-	-	-	-	-
18	-	-	-	-	-	-	-	-
19	-2.00	-1.00	-2.00	0.00	-10.00	-	-2.00	-
Weighted Mean	0.38	0.14	0.06	0.42	0.09	-0.55	0.07	0.06
Modal age	R17 IS	R18 ES	R19 FR	R20 NO	R21 NO	R22 PT	R23 IE	R24 FR
0	0.13	0.45	0.17	0.23	0.17	0.00	0.00	0.00
1	0.04	0.23	0.02	0.36	0.14	-0.21	-0.39	0.02
2	0.00	0.37	-0.12	0.41	0.21	-0.66	-0.22	-0.12
3	0.01	-0.09	-0.42	0.33	0.16	-0.93	-0.29	-0.16
4	0.02	-0.35	-1.26	0.51	0.50	-1.73	-1.23	-1.00
5	-0.18	-0.44	-1.82	0.57	0.47	-2.31	-1.42	-1.49
6	-0.12	-1.00	-2.64	0.39	0.14	-3.04	-1.86	-1.96
7	0.55	-0.91	-3.27	0.89	2.27	-3.36	-1.29	-2.18
8	-0.20	-1.70	-3.00	0.60	0.90	-4.20	-1.50	-1.70
9	0.38	-2.12	-3.00	0.86	1.25	-3.86	-0.60	-0.62
10	-0.25	-2.75	-4.25	0.33	1.25	-4.75	-	-2.00
11	0.00	-4.00	-7.00	0.00	1.00	-11.00	-	-1.00
12	-0.25	-4.25	-3.33	0.25	0.25	-6.00	-2.00	-0.75
13	0.00	-3.50	-4.50	0.00	0.00	-6.50	-1.00	-1.00
14	-	-	-	-	-	-	-	-
15	-	-	-	-	-	-	-	-

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Modal age	R17 IS	R18 ES	R19 FR		R20 NO	R21 N	10	R22 PT	R23 IE	R24 FR	
16	-	-	-		-	-		-	-	-	
17	-	-	-		-	-		-	-	-	
18	-	-	-		-	-		-	-	-	
19	0.00	-11.00	-9.0	0	-6.00	-1.00		-14.00	-	-3.00	
Weighted Mean	0.01	-0.28	-0.9	6	0.39	0.34		-1.45	-0.62	-0.60	
Modal age		R2	5 NO	R26	NO		R27	DK		all	
0		0.0	3	0.13			0.39			0.15	
1		0.0	0	0.04			0.16			0.10	
2		0.1	2	0.15			0.06			0.09	
3		0.0	7	0.10			-0.08	5		-0.04	
4		0.2	2	0.09			-0.24				
5		0.3	3	0.03			-0.77	,		-0.40	
6		0.0	7	-0.29)		-1.18	5		-0.69	
7		0.8	2	0.27	.27 -1.82					-0.41	
8		0.6	0	0.10)		-0.90			-0.68	
9		1.1	2	0.38		-1.25				-0.32	
10		0.5	0	0.25			-1.00)		-	
11		0.0	0	0.00		-5.00				-	
12		0.3	3	-1.00)		-1.25			-1.40	
13		1.0	0	0.00			-3.00)		-1.31	
14		-		-			-			-	
15		-		-			-			-	
16	-			-			-			-	
17	17			-			-			-	
18				-			-			-	
19			00	-5.00)		-1.00)	-		
Weighted Mean		0.1	7	0.05			-0.30)		-0.13	

Table 3.2. All areas and all the readers. Inter reader bias test. The Inter-reader bias test gives probability of bias between readers and with modal age = no sign of bias (p>0.05), *	= possibility
of bias (0.01 <p<0.05), (p<0.01).<="" *="certainty" bias="" of="" td=""><td></td></p<0.05),>	

Com- pari- son	R01 ES	R02 IE	R03 DK	R04 PT	R05 NL	R06 PT	R07 RU	R08 RU	R09 FO	R10 NO	R11 NO	R12 FO	R13 DE	R14 ES	R15 NO	R16 DE	R17 IS	R18 ES	R19 FR	R20 NO	R21 NO	R22 PT	R23 IE	R24 FR	R25 NO	R26 NO	R27 DK
R01 ES	-	**	-	**	**	-	**	-	**	-	-	**	**	**	-	*	-	**	**	**	**	*	*	-	*	-	*
R02 IE	**	-	*	**	*	*	**	**	**	**	-	**	**	*	-	**	-	-	*	**	*	*	*	*	**	-	**
R03 DK	-	*	-	**	*	**	**	**	*	**	**	*	*	*	**	*	**	-	-	*	*	*	*	**	*	*	*
R04 PT	**	**	**	-	**	**	**	**	**	-	-	**	*	**	-	-	-	*	**	**	**	*	**	*	-	-	*
R05 NL	**	*	*	**	-	*	**	**	*	**	-	*	**	*	-	-	**	*	*	*	*	*	*	*	**	-	**
R06 PT	-	*	**	**	*	-	**	**	*	**	**	*	**	*	**	**	**	-	**	*	*	*	*	**	**	**	*
R07 RU	**	**	**	**	**	**	-	*	**	-	**	**	-	**	**	**	**	*	*	**	**	*	**	**	-	**	**
R08 RU	-	**	**	**	**	**	*	-	**	-	**	**	-	**	*	**	**	*	**	**	**	*	**	**	-	-	**
R09 FO	**	**	*	**	*	*	**	**	-	-	-	*	**	*	-	-	-	**	*	*	*	*	*	*	**	-	**
R10 NO	-	**	**	-	**	**	-	-	-	-	**	-	*	**	**	*	**	**	*	**	**	*	**	**	*	**	-
R11 NO	-	-	**	-	-	**	**	**	-	**	-	*	*	**	*	*	**	**	*	**	-	*	**	**	**	*	-
R12 FO	**	**	*	**	*	*	**	**	*	-	*	-	**	*	-	-	**	**	*	*	**	*	*	*	-	-	**

R01 R02 R03 R08 R09 R10 R11 R12 R13 R14 R15 R18 R19 R20 R21 R22 R24 R25 R26 R27 Com-R04 R05 R06 R07 R16 R17 R23 pari-ES IE DK PT NL PT RU RU FO NO NO FO DE ES NO DE IS ES FR NO NO РТ IE FR NO NO DK son ** ** ** * * ** ** ** ** ** * ** ** ** R13 ** ** * * * * * -DE R14 ES ** ** * ** ** * ** ** * * * * ** ** ** * * ** * ** ** ** ** * * * ** * ** ** ** ** * ** ** ** * * * R15 . . ----NO R16 * ** * ** ** ** * * ** * * ** ** * ** * * ** * * DE ** ** ** ** ** ** ** ** ** ** * ** ** ** ** ** ** ** * * ** R17 IS -_ --** ** ** ** ** ** * R18 ES ** ** ** ** ** ** ** ** ** -R19 FR ** ** ** ** * * * * ** * * ** * * ** * * * ** * * * * * -R20 ** ** ** ** * ** ** * ** ** ** * ** * * ** ** * NO ** ** ** ** ** R21 ** ** NO R22 PT * * ** ** ** * R23 IE * ** ** ** ** * ** ** * ** ** * ** ** ** * * * ** * * * * * ** ** ** ** R24 FR -* ** * ** ** ** * * ** ** ** ** * * ** ** ** _ R25 * ** * ** ** ** * ** -* ** ** * ** ** * ** * ** ** ** --NO ** ** * ** * * ** ** * * ** ** * R26 * -** ---* -* * ---NO

R01 R02 R03 R04 R08 R09 R10 R11 R14 R15 R17 R18 R19 R20 R21 R24 Com-R05 R06 R07 R12 R13 R16 R22 R23 R25 R26 R27 pari-ES IE DK РТ NL РТ RU RU FO NO NO FO DE ES NO DE IS ES FR NO NO FR NO NO DK PT IE son R27 * ** * * ** * ** ** ** ** ** ** * ** ** ** * * -_ _ _ -. -_ -DK ** Modal -** ** ** ** * ** ** ** ** ** ** ** ** ** * ** ** * ** ** ** * age

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Figure 3.1. All areas and all the readers. The relative bias by modal age as estimated by all age readers combined.



Figure 3.2. All areas and all the readers. CV, PA (Percentage of agreement) and STDEV (standard deviation) are plotted against modal age.

In Figure 3.3. is represented the measurement (mm), as a distance from the center, of each annulus based on the classifications from the advanced readers on the otoliths by each ICES area. The otoliths from areas 27.2.a, 27.5.b.1, 27.6.b and 27.7.c were the ones presenting more differences in age ring identification between the advanced readers.



Figure 3.3. All areas and only the advanced readers. Plot of average distance from the centre to the winter rings for all advanced readers by ICES area. The boxes represent the median, upper and lower box boundaries of the interquartile range, whiskers represent the minimum and maximum values and the dots represent the outliers.

3.3 Northern areas pre-workshop results

Summary of statistics; Total number of samples (NSample), coefficient of variance (CV), percentage of agreement (PA) and average percentage error (APE) for all ages and readers from the Northern (All and Advanced).

NSample	cv	РА	APE	Readers
190	20%	69%	11%	All
190	20%	69%	11%	Advanced

Considering only the northern areas and only the age readers that usually read the otoliths from the northern areas, the relative bias was found to be minimal (-0.10), but for individual age-readers the relative bias varied from -0.91 to +0.36 (Table 3.3, Table 3.4 and Figure 3.4). This shows a significant over- and under-ageing of otoliths by age readers, and high bias was found both among experienced readers and trainees. For fish older than five years old there is a tendency of underestimating the ages, while overestimation seems to be the main problem among younger individuals. This is also evident on an individual basis where older fish were generally underaged compared to modal age, while there was variation in whether a reader under- or overestimated the ages of younger fish. The under/over-ageing signifies systematic miss-interpretation of growth structures within the otolith. There is also clear bias among readers compared to the I

modal age. The precision (CV), percentage of agreement and the standard deviation (STDEV) by modal age for all readers (Figure 3.5) reveals that the agreement decreases below 50% from the age 5 onwards.

Modal age	R01 IE	R02 DK	R03 NL	R04 RU	R05 RU	R06 FO	R07 NO	R08 NO
0	0.00	0.00	0.00	0.00	0.33	0.00	0.00	0.00
1	0.08	0.00	0.08	0.04	0.00	0.00	0.00	0.00
2	0.50	-0.10	0.43	0.07	-0.07	-0.10	-0.10	0.00
3	0.19	-0.40	0.09	-0.05	-0.18	0.21	0.12	0.09
4	0.48	-1.04	0.04	-0.40	-0.28	0.28	-0.08	0.00
5	0.29	-1.82	0.04	-0.62	-0.25	0.39	-0.21	0.04
6	0.46	-1.96	0.43	-0.71	-0.54	0.37	-0.26	-0.12
7	-0.17	-2.33	0.33	-0.67	-0.50	1.00	-0.50	0.50
8	0.64	-1.90	0.36	-0.18	0.09	0.45	-0.73	0.64
9	0.80	-1.20	0.80	-0.40	0.00	1.60	-0.20	0.25
10	1.60	-1.00	0.60	-0.40	-0.40	-0.60	0.20	0.80
11	1.50	-2.00	0.00	-0.50	-0.50	1.50	-0.50	0.00
12	0.25	-2.75	-0.50	-1.25	-1.25	0.00	-0.25	0.25
13	0.00	-0.50	-1.00	-2.00	0.50	0.50	-0.50	0.50
14	-	-	-	-	-	-	-	-
15	-	-	-	-	-	-	-	-
16	-	-	-	-	-	-	-	-
17	-	-	-	-	-	-	-	-
18	-	-	-	-	-	-	-	-
19	0.00	-3.00	-1.00	-5.00	0.00	-2.00	-2.00	-1.00
Weighted Mean	0.36	-0.96	0.18	-0.31	-0.21	0.24	-0.12	0.08
Modal age	R09 FO	R10 DE	R11 DE	R12 NO	R13 NO	R14 NO	R15 IS	R16 IE
0	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
1	0.08	0.04	0.08	0.04	0.08	0.00	0.00	-0.42

Table 3.3 Northern areas with only northern readers. The relative bias (as the difference between the mean and modal age) per modal age and reader is presented, as well as the weighted mean relative bias per reader and the relative bias per modal age for all readers combined.

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Modal age	R09 FO	R10 DE	R11	DE	R12 NO	R13 NO	R14 NO	R15 IS	R16 IE
2	0.30	0.40	0.37		0.03	0.24	0.13	-0.07	-0.13
3	0.12	0.19	0.07		0.05	0.21	0.09	-0.07	-0.32
4	0.00	-0.28	-0.36	5	-0.08	0.00	0.12	-0.40	-1.24
5	0.00	-0.11	-0.32	2	0.00	0.14	0.19	-0.32	-1.73
6	0.29	-0.04	-0.30)	-0.19	0.24	-0.04	-0.27	-2.25
7	0.50	-0.17	-0.17	7	-0.17	0.60	1.50	0.00	-2.20
8	1.18	-0.09	-0.45	5	0.55	0.50	1.55	-0.09	-1.29
9	1.00	-0.60	-0.80)	-0.40	0.50	0.80	-0.20	-0.67
10	-0.40	-0.80	-1.00)	0.00	0.25	1.00	-0.40	-2.00
11	1.00	0.00	0.00		0.00	0.50	1.00	0.00	-2.00
12	0.00	-1.25	-1.50)	-0.50	0.25	0.25	-0.25	-2.00
13	0.50	-2.00	-2.00)	-0.50	0.00	0.00	0.00	-1.00
14	-	-	-		-	-	-	-	-
15	-	-	-		-	-	-	-	-
16	-	-	-		-	-	-	-	-
17	-	-	-		-	-	-	-	-
18	-	-	-		-	-	-	-	-
19	0.00	-10.00	-		-2.00	-6.00	-1.00	0.00	-
Weighted Mean	0.21	-0.06	-0.15	5	-0.02	0.17	0.23	-0.16	-0.91
Modal age		R17	NO	R18	NO	R19	DK	all	
0		0.00		0.00		0.3	3	0.04	1
1		0.00)	0.00)	0.0	8	0.01	L
2		0.13		0.10)	0.0	7	0.12	2
3		0.04	ļ	0.04	Ļ	-0.1	2	0.02	2
4		0.04		-0.04	4	-0.6	54	-0.2	0
5		-0.0	4	-0.1	4	-1.2	25	-0.3	0
6		0.14		-0.4	1	-1.1	4	-0.3	3
7		0.00)	-0.3	3	-1.6	57	-0.2	3
8		0.91		0.00)	-1.3	86	0.04	1

Modal age	R17 NO	R18 NO	R19 DK	all
9	0.80	-0.20	-1.60	0.01
10	0.40	0.20	-1.20	-0.17
11	1.00	0.00	-3.00	-0.11
12	0.33	-1.00	-1.25	-0.65
13	1.00	0.00	-3.00	-0.50
14	-	-	-	-
15	-	-	-	-
16	-	-	-	-
17	-	-	-	-
18	-	-	-	-
19	-3.00	-5.00	-1.00	-
Weighted Mean	0.12	-0.10	-0.61	-0.10

Table 3.4. Northern areas with only northern readers. Inter reader bias test. The Inter-reader bias test gives probability of bias between readers and with modal age. - = no sign of bias (p>0.05), * = possibility of bias (0.01< p<0.05), * = certainty of bias (p<0.01).

Com- pari- son	RO 1 IE	R0 2 DK	RO 3 NL	RO 4 RU	RO 5 RU	R0 6 FO	R0 7 NO	R0 8 NO	R0 9 FO	R1 0 DE	R1 1 DE	R1 2 NO	R1 3 NO	R1 4 NO	R1 5 IS	R1 6 IE	R1 7 NO	R1 8 NO	R1 9 DK
R01 IE	-	*	**	**	**	**	-	*	**	-	*	-	**	**	**	**	**	**	**
R02 DK	*	-	**	**	**	**	**	**	**	**	**	**	**	**	**	*	**	**	-
R03 NL	**	**	-	**	**	**	-	**	*	**	-	-	**	**	*	**	**	-	**
R04 RU	**	**	**	-	**	**	**	-	**	-	**	-	**	**	**	**	**	*	**
R05 RU	**	**	**	**	-	-	**	-	*	**	**	*	*	-	**	**	-	**	-
R06 FO	**	**	**	**	-	-	-	**	**	*	-	*	*	**	**	**	**	-	**
R07 NO	-	**	-	**	**	-	-	*	-	**	*	**	*	-	**	**	**	**	-
R08 NO	*	**	**	-	-	**	*	-	**	*	**	**	**	**	*	**	**	**	**

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Com- pari- son	RO 1 IE	R0 2 DK	RO 3 NL	RO 4 RU	RO 5 RU	R0 6 FO	R0 7 NO	R0 8 NO	R0 9 FO	R1 0 DE	R1 1 DE	R1 2 NO	R1 3 NO	R1 4 NO	R1 5 IS	R1 6 IE	R1 7 NO	R1 8 NO	R1 9 DK
R09 FO	**	**	*	**	*	**	-	**	-	**	-	*	*	**	-	**	**	*	**
R10 DE	-	**	**	-	**	*	**	*	**	-	-	*	**	**	**	**	**	**	**
R11 DE	*	**	-	**	**	-	*	**	-	-	-	**	-	**	**	**	**	**	-
R12 NO	-	**	-	-	*	*	**	**	*	*	**	-	-	*	**	**	**	**	-
R13 NO	**	**	**	**	*	*	*	**	*	**	-	-	-	**	**	**	**	-	**
R14 NO	**	**	**	**	-	**	-	**	**	**	**	*	**	-	-	**	**	-	**
R15 IS	**	**	*	**	**	**	**	*	-	**	**	**	**	-	-	**	-	**	**
R16 IE	**	*	**	**	**	**	**	**	**	**	**	**	**	**	**	-	**	**	-
R17 NO	**	**	**	**	-	**	**	**	**	**	**	**	**	**	-	**	-	-	**
R18 NO	**	**	-	*	**	-	**	**	*	**	**	**	-	-	**	**	-	-	-
R19 DK	**	-	**	**	-	**	-	**	**	**	-	-	**	**	**	-	**	-	-
Moda I age	**	**	-	*	-	-	**	**	*	*	**	**	*	-	-	**	**	**	**



Figure 3.4. Northern areas with only northern readers. The relative bias by modal age as estimated by all age readers combined. CV, PA (Percentage of agreement) and STDEV (standard deviation) are plotted against modal age.



Figure 3.5. All areas and all the readers. CV, PA (Percentage of agreement) and STDEV (standard deviation) are plotted against modal age.

In Figure 3.6. is represented the measurement (mm), as a distance from the center, of each annulus based on the classifications from the advanced northern readers on the otoliths by each ICES area only considered the Northern areas of the stock distribution. The otoliths from areas 27.2.a, 27.5.b.1, 27.6.b and 27.7.c were the ones presenting more differences in age ring identification between the advanced readers.

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Figure 3.6. Northern areas with only advanced northern readers. Plot of average distance from the center to the winter rings for all advanced readers by strata. The boxes represent the median, upper and lower box boundaries of the interquartile range, whiskers represent the minimum and maximum values and the dots represent the outliers.

3.4 Southern areas pre-workshop results

Summary of statistics: Total number of samples (NSample), coefficient of variance (CV), percentage of agreement (PA) and average percentage error (APE) for all ages and readers from the Southern (All and Advanced).

NSample	cv	РА	APE	Readers
217	29%	72%	18%	All
217	24%	79%	14%	Advanced

Considering only the southern areas and only the age readers that usually read the otoliths from the southern, the relative bias was found to be minimal (-0.1), but for individual age-readers the relative bias varied from -0.1 to +0.6 (Table 3.5, Table 3.6, and Figure 3.7). This shows a significant over- and under-ageing of otoliths by age readers, and high bias was found both among experienced readers and trainees. For fish older than five there is a tendency of underestimating the ages, while overestimation seems to be the main problem among younger individuals. This is also evident on an individual basis where older fish were generally under-aged compared to modal age, while there was variation in whether a reader under- or overestimated the ages of younger fish. The under/over-ageing signifies systematic miss-interpretation of growth structures within the otolith. There is also clear bias among readers compared to the modal age. The precision (CV), percentage of agreement and the standard deviation (STDEV) by modal age for all readers (Figure 3.8) reveals that the agreement decreases below 50% from the age 5 onwards.

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Modal age	R01 ES	R02 PT	R03 PT	R04 ES	R05 ES	R06 FR	R07 PT	R08 FR
0	0.1	0.0	0.3	0.0	0.5	0.2	0.0	0.0
1	0.2	0.2	0.1	0.1	0.6	0.0	0.0	0.1
2	0.2	0.2	-0.1	-0.2	0.7	-0.1	-0.6	-0.1
3	0.2	0.2	-0.2	-0.4	0.2	-0.3	-0.9	0.0
4	0.4	0.2	-0.4	-0.6	0.6	-1.0	-1.0	0.0
5	-0.2	0.4	-0.5	-0.2	0.4	-1.1	-1.5	0.1
6	0.3	0.7	-0.3	0.0	0.7	-1.3	-2.3	0.3
7	-0.5	2.0	0.0	-1.5	0.0	-1.5	-3.0	1.0
Weighted Mean	0.2	0.2	-0.1	-0.2	0.5	-0.3	-0.6	0.0
Modal age		all						
0		0.1						
1		0.2						
2		0.0						
3		-0.1						
4		-0.2						
5		-0.3						
6		-0.2						
7		-0.4						
Weighted Mean		0.0						

Table 3.5. Southern areas with only southern readers. The relative bias (as the difference between the mean and modal age) per modal age and reader is presented, as well as the weighted mean relative bias per reader and the relative bias per modal age for all readers combined.

Comparison	R01 ES	R02 PT	R03 PT	R04 ES	R05 ES	R06 FR	R07 PT	R08 FR
R01 ES	-	**	**	**	-	**	**	-
R02 PT	**	-	-	**	*	**	**	-
R03 PT	**	-	-	**	**	-	**	**
R04 ES	**	**	**	-	**	**	**	-
R05 ES	-	*	**	**	-	**	**	**
R06 FR	**	**	-	**	**	-	**	-
R07 PT	**	**	**	**	**	**	-	**
R08 FR	-	-	**	-	**	-	**	-
Modal age	*	**	**	*	**	-	**	**

Table 3.6. Southern areas with only southern readers. Inter reader bias test. The Inter-reader bias test gives probability of bias between readers and with modal age. - = no sign of bias (p>0.05), * = possibility of bias (0.01< p<0.05), * = certainty of bias (p<0.01).



Figure 3.7. Southern areas with only southern readers. The relative bias by modal age as estimated by all age readers combined.

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Figure 3.8. Southern areas with only southern readers. CV, PA (Percentage of agreement) and STDEV (standard deviation) are plotted against modal age.

Figure 3.9 represented the measurement (mm), as a distance from the center, of each annulus based on the classifications from the advanced southern readers on the otoliths by each ICES area only considered the Southern areas of the stock distribution.



Figure 3.9. Southern areas with only advanced southern readers. Plot of average distance from the center to the winter rings for all advanced readers by strata. The boxes represent the median, upper and lower box boundaries of the interquartile range, whiskers represent the minimum and maximum values and the dots represent the outliers.
4 Update the guidelines and common age reading criteria (ToR c and d)

4.1 Plenary discussion of otoliths

A selection of difficult otoliths was discussed in plenary using SmartDots. It was early established that there was a clear difference in interpretation of the rings between readers of the southern areas and readers of the northern areas. It is expected that these differences are related to the growth patterns of the fish from the different areas. Around the Iberian Coast the water is much warmer than at north of the British Isles, and higher growth should be expected.

The variation in ages given by the readers was often very high. This was mainly because readers of both stock areas had read all otoliths. When excluding the readers of the opposite stock areas, it was easier to discuss the issues of the otolith zonation. However, it was also very important to include the readers of the opposite areas in the discussions of interpretation of the otolith zonation, to ensure all possible errors were discussed.

Because this was online, it was not possible for the participants to annotate the age rings in the otolith during the discussions, as they would normally do. This made the discussions a bit more difficult. But by using the image annotations in SmartDots it was fundamental to go through the otoliths during the plenary session and to identify the differences on age classifications between the participants.

4.2 Issues

It was not always possible to get to an agreement, especially when the endpoints of the inner zone(s) where difficult to see. However, regarding the disagreement about the pattern of the zonation in most of the cases after discussion it was rather easy to get to an agreement.

First ring interpretation issues still remain. There are clear differences in the age 1 ring length between the southern and the northern areas of the blue whiting stock distribution (see details in Section 2.1.1 and Annex 5). The data analysis from the 2020 pre-workshop exchange revealed that the age 1 ring length seems to increase in size as the fish gets older and this was observed in all the main areas of this species distribution, also observed in the otoliths from the Mediterranean (Figure 4.1).

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Figure 4.1. Boxplot of the first age ring length (mm) by otolith total length (mm) across the different modal ages and for the main three areas: Mediterranean, Northern and Southern areas. The dashed blue lines represent the limits defined for the Bailey's zone, between 8.00 and 9.33 mm.

The data presented in figure 4.1. includes all the otoliths from the 2020 pre-workshop exchange, with a percentage of agreement on the modal age between 26 and 100%. To avoid a misinterpretation of the first age ring due to including difficult to read or unreadable otoliths (AQ2 and AQ3), the data by area was re-analysed and only included the results from the otoliths in which a 60% or higher percentage of agreement between the readers was achieved (Figure 4.2).

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Figure 4.2. Boxplot of the first age ring length (mm) by fish total length (mm) across the different modal ages (0-12) and for the different ICES areas. The dashed blue lines represent the limits defined for the Bailey's zone, between 8.00 and 9.33 mm.

In older otoliths the first ring seems wider and readers make assumptions about the possible existence of an inner first ring that are not visible (Figure 4.3. and Figure 4.5.). A possibility to check if there are an inner ring is to polish the otoliths. During the workshop it was attempted to validate the assumptions of the existence of a non-visible inner ring. For this, some selected otoliths have been polished (Figure 4.4. and Figure 4.6.).



Figure 4.3. Otolith with the first visible inner ring measuring 12.08 mm (Fish ID: FO_54).



Figure 4.4. Otolith polished without any other visible inner ring measuring less than 12.08 mm (Fish ID: FO_54).



Figure 4.5. Otolith with the first visible inner ring measuring 13.07 mm (Fish ID: IRL_21).



Figure 4.6. Otolith polished with a visible inner ring measuring less than 13.07 mm (Fish ID: IRL_21).

The polishing technique showed to be helpful, to clarify the existence of a non-visible inner ring, when the measurement of the first visible age ring is higher than the expected value of 9.33 mm.

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4.3 Clarify interpretation of annual growth rings (1-3)

See the guidelines section for clarification on annual growth rings interpretation (Section 4.5).

4.4 WKARBLUE3 age reading exercise

During the workshop a small age intercalibration exercise was performed using SmartDots (EventID 347), with a total of 55 otoliths (33 from the northern areas and 22 from the southern areas). On this small exercise have participated 11 readers from the northern areas (9 of those were advanced readers) and 5 from the southern areas (2 of those were advanced readers). The results have been analysed and presented by area and only included the readers that are used to read the otoliths from those areas (Northern areas – Section 4.4.1; Southern areas – Section 4.4.2).

4.4.1 Northern areas

In this section an overview of the samples from the workshop intercalibration event are described (Table 4.1) and the results presented (Table 4.2, 4.3, 4.4 and Figures 4.7, 4.8 and 4.9).

Year	ICES area	Quarter	Number of samples	Modal age range	Length range
2009	27.4.a	4	3	5-6	290-315 mm
2012	27.2.b	2	2	1-8	200-365 mm
2014	27.4.a	4	2	2-3	235-265 mm
2017	27.2.a	2	2	4-9	270-320 mm
2017	27.2.a	3	5	2-11	270-360 mm
2017	27.2.a	4	2	1-5	240-310 mm
2017	27.5.a	2	1	8	350 mm
2017	27.5.a	4	1	0	200 mm
2017	27.5.b	2	1	9	320 mm
2017	27.5.b.1	1	2	3-5	260-300 mm
2017	27.5.b.1	2	2	1-3	200-240 mm
2017	27.5.b.1	4	1	0	210 mm
2017	27.6.a	2	1	10	430 mm
2017	27.6.a	3	3	2-5	250-340 mm
2017	27.6.b	1	4	3-10	240-330 mm
2017	27.7.c	1	1	12	320 mm

 Table 4.1. Northern areas. Overview of samples (n=33) used for the exchange EventID 347.

NSample	cv	РА	APE	Readers
33	17 %	69 %	12 %	All
33	17 %	71 %	11 %	Advanced

Table 4.2. Northern areas. Summary of statistics; Total number of samples (NSample), coefficient of variance (CV), percentage of agreement (PA) and average percentage error (APE) for all ages and readers (All and Advanced).

Table 4.3. Northern areas. The relative bias (as the difference between the mean and modal age) per modal age and reader is presented, as well as the weighted mean relative bias per reader and the relative bias per modal age for all readers combined.

Modal age	R01 IE	R02 DK	R04	NL	R06 FO	R07 NO	R08 FO	R10 DE	R11 NO
0	0.0	0.00	0.50		0.0	0.0	0.0	0.00	0.00
1	0.3	0.33	0.00		0.0	0.0	0.7	-0.33	0.00
2	0.7	0.33	0.67		0.0	0.3	0.7	0.33	0.33
3	0.8	0.50	0.25		-0.2	0.0	0.2	0.25	0.00
4	0.3	-0.33	-0.33	}	-0.7	0.0	-0.5	-0.33	0.00
5	0.4	-0.25	-0.20)	-0.2	-0.2	0.4	0.00	0.00
6	0.3	-0.50	0.33		-0.7	0.0	0.0	0.00	0.00
7	0.5	-1.00	0.50		0.0	0.0	0.0	0.00	0.50
8	0.0	-1.50	0.00		0.5	0.5	0.5	0.00	0.00
9	6.0	0.00	0.00		-1.0	0.5	0.0	0.50	2.00
10	0.0	-2.00	0.00		-1.0	-0.5	0.0	-1.00	0.00
11	1.0	-	0.00		0.0	1.0	-1.0	-	2.00
12	0.0	-	-		-4.0	0.0	-3.0	-2.00	-
Weighted Mean	0.7	-0.22	0.12		-0.4	0.1	0.1	-0.09	0.20
Modal age		R12	IS	R15	IE	R18	NL	all	
0		0.00		0.0		0.0		0.05	
1		0.00		0.3		0.3		0.15	
2		0.00		0.7		0.0		0.36	
3		0.50		0.0		0.0		0.20	
4		0.00		0.0		-0.3		-0.20)

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Modal age	R12 IS	R15 IE	R18 NL	all
5	0.00	0.6	0.0	0.05
6	0.00	0.3	0.0	-0.02
7	1.00	0.0	0.0	0.14
8	0.00	1.0	0.0	0.09
9	0.00	0.0	0.0	0.73
10	0.00	-0.5	0.0	-0.45
11	2.00	0.0	-2.0	-
12	-	-4.0	-1.0	-
Weighted Mean	0.19	0.1	-0.1	0.09

Table 4.4. Northern areas. Inter reader bias test. The Inter-reader bias test gives probability of bias between readers and with modal age. - = no sign of bias (p>0.05), * = possibility of bias (0.01<p<0.05), * = certainty of bias (p<0.01).

Compari- son	R01 IE	R02 DK	R04 NL	R06 FO	R07 NO	R08 FO	R10 DE	R11 NO	R12 IS	R15 IE	R18 NL
R01 IE	-	-	-	**	*	-	**	-	-	-	*
R02 DK	-	-	*	-	**	-	**	-	-	-	**
R04 NL	-	*	-	-	*	**	**	**	*	-	*
R06 FO	**	-	-	-	*	-	-	*	*	*	-
R07 NO	*	**	*	*	-	-	-	-	-	-	*
R08 FO	-	-	**	-	-	-	-	*	**	**	-
R10 DE	**	**	**	-	-	-	-	-	-	-	**
R11 NO	-	-	**	*	-	*	-	-	*	-	-
R12 IS	-	-	*	*	-	**	-	*	-	*	*
R15 IE	-	-	-	*	-	**	-	-	*	-	-
R18 NL	*	**	*	-	*	-	**	-	*	-	-
Modal age	*	-	-	-	*	-	-	-	-	-	**



Figure 4.7. Northern areas. Plot of average distance from the center to the winter rings for all readers. The boxes represent the median, upper and lower box boundaries of the interquartile range, whiskers represent the minimum and maximum values and the dots represent the outliers. Note: it is only possible to present the data plots by all the combined ICES northern areas, due to small sample size from each area.



Figure 4.8. Northern areas.CV, PA and (STDEV (standard deviation) are plotted against modal age.



Figure 4.9. Northern areas. The relative bias by modal age as estimated by all age readers combined.

4.4.2 Southern areas

In this section an overview of the samples from the workshop intercalibration event are described (Table 4.5) and the results presented (Table 4.6, 4.7, 4.8 and Figures 4.10, 4.11 and 4.12).

Year	ICES area	Quarter	Number of samples	Modal age range	Length range
2017	27.8.c	1	2	1-3	180-215 mm
2017	27.8.c	2	4	3-7	265-350 mm
2017	27.8.c	3	4	1-10	245-320 mm
2017	27.8.c	4	4	3-7	250-370 mm
2017	27.9.a	1	2	2-7	185-300 mm
2017	27.9.a	2	1	4	265 mm
2017	27.9.a	3	2	2-4	295 mm
2017	27.9.a	4	3	0-5	195-315 mm

Table 4.5. Southern areas. Overview of samples (n= 22) used for the exchange EventID 347.

Table 4.6. Southern areas. Summary of statistics; Total number of samples (NSample), coefficient of variance (CV), percentage of agreement (PA) and average percentage error (APE) for all ages and readers (All and Advanced).

NSample	CV	РА	APE	Readers
22	23%	67%	15%	All
22	11%	78%	9%	Advanced

Modal age	R03 PT	R05 PT	R14 FR	R16 FR	R17 PT	all
0	0.00	0.00	0.00	0.00	0.00	0.00
1	0.00	0.00	1.00	0.00	-0.50	0.10
2	0.50	0.00	0.00	-0.50	0.50	0.10
3	0.00	-0.33	0.00	-0.33	0.00	-0.13
4	0.25	-0.25	-0.50	0.00	0.25	-0.05
5	1.00	-0.33	0.00	0.33	0.33	0.27
6	0.00	-1.00	0.00	0.00	-2.00	-0.60
7	0.50	-1.00	-1.25	-1.00	-0.75	-0.70
8	0.00	0.00	-2.00	-1.00	-2.00	-1.00
9	-	-	-	-	-	-
10	0.00	0.00	-2.00	-1.00	-3.00	-1.20
Weighted Mean	0.33	-0.35	-0.41	-0.32	-0.36	-0.23

Table 4.7. Southern areas. The relative bias (as the difference between the mean and modal age) per modal age and reader is presented, as well as the weighted mean relative bias per reader and the relative bias per modal age for all readers combined.

Table 4.8. Southern areas. Inter reader bias test. The Inter-reader bias test gives probability of bias between readers and with modal age. - = no sign of bias (p>0.05), * = possibility of bias (0.01<p<0.05), * = certainty of bias (p<0.01).

Comparison	R03 PT	R05 PT	R14 FR	R16 FR	R17 PT
R03 PT	-	**	*	**	-
R05 PT	**	-	-	**	-
R14 FR	*	-	-	-	-
R16 FR	**	**	-	-	-
R17 PT	-	-	-	-	-
Modal age	*	*	-	-	-

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Figure 4.10. Southern areas. Plot of average distance from the center to the winter rings for all readers. The boxes represent the median, upper and lower box boundaries of the interquartile range, whiskers represent the minimum and maximum values and the dots represent the outliers. Note: it is only possible to present the data plots by all the combined ICES southern areas, due to small sample size from each area.



Figure 4.11. Southern areas. CV, PA and (STDEV (standard deviation) are plotted against modal age.



Figure 4.12. Southern areas. The relative bias by modal age as estimated by all age readers combined.

4.5 Updated guidelines

4.5.1 General guidelines for age determination. Ageing manual

Quick guidelines	
Preparation	Soak for 24 hours before reading using reflected light.
Identification of nucleus	Beware of the Bailey's zone, and keep in mind the possible size of the first growth zone.
Issues	False or split rings occur. It is important to beware of a reasonable growth pattern in the otolith.
Interpretation of the edge	Growth varies with age, and the two schemes made should always be consulted.
Stock areas	Growth pattern is different between specimens from the northen and southern areas of the stock.
Age plus-group	The age plus-group considered on stock assessment is age 10.
Sexual dimorphism	Females grow faster than males, especially after the age of first maturity.
Reference collection	Use reference collection to calibrate before reading for assessment.

- Blue whiting **whole otoliths** must be soaked in water 24 hours before reading (only if they are dried after sampling. Otoliths read straight after collection do not need to be soaked for 24 hours prior to reading). The otoliths should not be soaked in water for more than 48 hours each time, as it possibly could affect the ring structure due to the composition of freshwater (Anon.1992). No other manipulation is needed. It is, however, important to age the otolith shortly after sampling, as the otoliths are clearest then.
- Whole otoliths must be read in water over a black surface, using reflected light.

- **Magnification** of images should always be the same (x 0.64), and a measurement bar needs to be included in all images of blue whiting. This is very important in order to correctly determine the inner zone vs Bailey's zone.
- Be aware of which **end of the otolith** is read from. Read from centre to pointy edge, and read on the dorsal side (upside when placed in the inner ear of the fish) (Härkönen, 1986). Often the rings can only be followed from the centre towards one end of the otolith, while they will merge very close to the pointy edge at the other end (Figure 4.13).

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Figure 4.13 The red dots indicate the correct direction to read. However, it is important to follow the rings along the side as well. The lines can clearly be followed on the side with the orange dots, but when reading along the blue dots/transect the age goes from 5 to 4 years old. We regard the red circle to be within the area it should be possible to follow the zones to call them annual zones (orange dots).

- Have in mind which area the fish is captured in. There might be differences in growth pattern between areas, especially for fish captured in the northern areas and the southern areas of the stock (Figure 4.14) (See sections 4.6 and 4.7 for clarifications).
- The age is given by the number of translucent (winter) zones in the otolith (Jakupsstovu, 1979).
- Correct **identification of annulus** can be induced by measuring the size of the inner ring. It will thereby be possible to avoid including the Bailey's zone (Bailey, 1970) as the first annual ring. The Bailey's zone is considered to be less than 8 mm in diameter. The first ring can be invisible or difficult to see in larger otoliths. It can be a good idea to have a 0 or 1-year-old otolith available when reading to easily compare the size of the inner ring.
- **Growth pattern** of the otolith zonation should be reasonable. It is expected that the inner zones will be wide, and the zones get more and more narrow towards the edge (Figure 4.15). It is, however, important to look at a group of fish from the same area of the stock. If several fish with the same measurements show the same pattern, it could be a biological reason to dismiss a normal growth pattern.



Figure 4.14. The white shaded area indicates the position of the transition zone between the putative northern and southern stocks proposed by Skogen et al. (1999). (Figure adapted from ICES Ecoregions including ICES Statistical Areas, ices.dk Dec. 2017).

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Figure 4.15. The width of the zones should be expected to become smaller with distance from the nucleus. Variation can be expected due to variation in feeding, spawning etc.

- **False rings** are a common issue in blue whiting otoliths. When counting the true annual rings to age the fish, it is important to look at the entire structure of the otolith and follow the sequence of yearly growth. The yearly growth zone increments will most often decrease as the fish get older. When small growth zones are followed by bigger ones, these should be considered as false rings. However, sometimes ring thickness varies within the otolith, and a translucent zone may appear very thin, but is in good sequence, which could be a short winter period and not a false ring.
- A particular case of false rings are the **split rings** (double rings). In many cases they can be easily identified because they merge when you try to follow them around the otolith. It is important to follow the rings as far as possible to the side of the otolith. Zooming out will reduce the possibility of counting split rings. When in doubt check for expected growth pattern (Figure 4.16).



Figure 4.16 The blue light dots of the image show the split rings. Only the red dots should be counted as one year.

• Interpretation of the edge is determined by the time of capture of the fish. By an international convention (Williams and Bedford, 1974), the birthdate of fish has been assigned as January 1st, regardless of hatch date. A fish age-reader must assign a fish to its proper age according to the date it was caught with reference to this birthdate. Criterion of birthday on January 1st must be used to determine when a hyaline ring in the edge must be counted. Growth of immature fish vary from that of adults, as they may feed for a much longer period, and the opaque zone may therefore start forming already in March. Aging of a fish with an opaque edge present will therefore depend on maturity. Otoliths with translucent edge, sampled from the first half of the year, are aged by counting all translucent annuli, including the edge. Second half of the year, are aged by ignoring a translucent edge if present. This 'translucent edge' is the onset of the winter ring. This onset will also vary with time and by geographic location. This scheme must be clarified and validated in the future. This modified scheme is made for blue whiting during the workshop in 2013 (ICES 2013) using the figure from WKACM2 (Figure 4.17).



Figure 4.17. Modified scheme for Blue whiting.

• Blue whiting age readers **should avoid otoliths classified as unreadable** or very difficult to interpret (AQ3) according to the following 3-point scale of age reading quality that WKNARC (ICES 2011) (see details in Section 8) recommends to be used by all age readers who provide age data for stock assessments:

AQ1: Easy to age with high precision.

AQ2: Difficult to age with acceptable precision.

AQ3: Unreadable or very difficult to age with acceptable precision.

- **Reference Collections** should be used as a valuable tool to maintain the accuracy of readers over time. When readers are not reading blue whiting otoliths continuously throughout the year, the reference collection should be used to calibrate oneself before reading for assessment.
- **Training Collection** should be prepared to be used as a tool on training new readers. Reference Collections should also be used as a training collection.
- Sexual dimorphism is present in blue whiting, females grow faster than males, thus are younger in general at similar length to male fish (ICES 2005). This can be observed in the ring patterns in the otoliths with male fish tending to have smaller increments due to slower growth and is especially seen in fish after first maturity (Figure 4.18). Therefore, knowledge of the sex of the fish may be used as additional factor when ageing.

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Figure 4.18. Otolith length (mm) by fish age and by sex: NA (red), female (blue), male (green) and undefined (purple).

4.5.2 Additional supporting information on age reading

- The ring called Bailey's zone was first identified by Roger Bailey (Bailey, 1970); In samples of small blue whiting taken by small-meshed trawl in June 1967, Bailey found two distinct modal size groups, one of them around 8-9 cm, the other 13-15 cm. with no clearly defined winter growth rings in the otoliths. However, he considered that to attain a length of 13-15 cm by late June in these areas was unlikely and therefore proposed that, whereas the smaller size group may have been spawned that year, the larger ones were more likely to be 1-year-old. He also found that most of the otoliths of the second modal group (13-15 cm) showed a very indistinct ring when viewed in transverse section and this, he thought, may have been a weakly-developed first winter growth check. That zone was called thereafter Bailey's Zone.
- Jakupsstovu (1979) suggested that this zone may be associated with a change of habit or depth; in this case it would be equivalent to the "Bowers Zone" found in whiting otoliths (Gambell and Messtorff, 1964). This first zone is formed when the fish are 4-10 cm in length. Bailey (1982) concludes that it is difficult to explain how the youngest age group of blue whiting could have remained totally unobserved throughout their first winter and spring, especially if one considers their undoubted abundance. On the face of it, therefore, Jakupsstovu's interpretation seems more credible, and for the sake of consistency it is probably wisest at present to follow Jakupsstovu's (1979) interpretation in which the age is given by the number of winter rings on the otolith.
- Mature fish begin to grow later in the year than immature by reason of using the energy resource for gonad maturation cycle vs. using it to the somatic growth only by young fish. Blue whiting may mature already around age 1 (ICES 2013a). Thus, when possible, maturity stage should be used as an additional indicator for aging of fish caught during the spawning season.

- **Growth** begins when fish start feeding after the winter period/spawning and finish feeding after the accumulation of enough food reserves. In the last quarter, growth is finished due to enough energy resource for next spawning period, and the next winter ring has started to form. Therefore, stomach fullness can be used as an additional indicator for reading.
- Blue whiting has a wide distribution and a complicated life cycle in Atlantic waters. It can be reflected in all phases of the fish growth, and consequently in the otolith. The distribution is reflected in the landings, as shown in the map that the WGWIDE includes each year (Figure 4.19). The map also shows spawning concentrations west of the British Isles (Porcupine and Rockall Bank). That is during winter, where they don't feed and instead spend energy on spawning, and in the pre-spawning and post-spawning migrations. Since in the winter season, there is almost no otolith calcification, on the otolith a translucent ring is formed. After that, eggs and larvae drift mainly northward but also partly southward, recruiting to the nursery areas of the north (mainly Norwegian Sea) and the south (mainly Biscay Bay). At the same time, adult blue whiting migrates to the feeding grounds (in the same areas as the nurseries). They spread all around the Norwegian Sea, and part of the stock distribution is so scattered that it can't be detected by the fisherman or the surveys. That is shown in the 3rd and 4th quarters maps. In the feeding area the fish grow and in the otolith a wide opaque ring is marked.
- Another factor that affects the otolith growth is the **strength of recruitment**. Blue whiting stock alternates from periods of high recruitment regime to others of low recruitment. That affects the otolith growth and any other density-dependent characteristic.
- Growth is different between northern and southern areas of the stock. Fish from the northern areas has a slower growth than fish in warmer areas. Older males tend to be more difficult to age compared to females due to thinner and narrower rings.

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Figure 4.19. Blue whiting total catches (ICES estimates) in 2020 by quarter and ICES statistical rectangles. The catches on the map are based on logbook data and constitute 98.9 % of the ICES estimated catches. The total catches and percentages shown on each panel are also based on logbook data, and therefore deviate slightly from the ICES estimated catches pr. quarter. The 200 m and 1000 m depth contours are indicated in blue (ICES. 2021).

4.6 Differences between stock areas

There are clear differences in otolith growth patterns between the two stock areas of blue whiting. The age reading criteria are the same, but there are details one need to be explicit about when ageing the fish from different areas.

4.6.1 Northern areas

• Northern age readers can often encounter a split ring in the second year of growth (Figure 4.20)



Figure 4.20. Blue whiting otoliths with a split ring identified in the second year of growth (yellow arrow).

• As the fish get older the otolith becomes denser and can hide the 1st and even the 2nd rings, so measurements are needed to identify these rings and should not be avoided (Figure 4.21). Place a younger otolith with clear rings beside the difficult otolith can give you guidance on where the first and second ring could be hiding. It can be difficult to measure as the rings at the posterior end is often invisible.



Figure 4.21. Blue whiting otoliths of an older fish showing a dense aspect with the possible hide of the 1st and maybe the 2nd rings.

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• A male and female fish of equal age can have big difference in length, the female being the biggest (Figure 4.22). Due to the different size at age, males can be more difficult to age, as they have more narrow zones.



Figure 4.22. Blue whiting otoliths of female with 430 mm total length (ICE 32-11) and of a male with 330 mm total length (ICE 29-2), both with 9 years old.

- We expect the winter ring to be formed already in Q4 and should not be counted until following January.
- Even in summer some otoliths can show very little opaque zone due to late start to feeding in northern waters (Figure 4.23)



Figure 4.23. Blue whiting otoliths from fish captured in the summer (FO 54 from August and ICE 41-18 from June) presenting a small opaque zone.

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• Some fish start feeding in early spring and will show a thin opaque zone already starting to form, so adding another year's growth at the edge should not be included if this is found (Figure 4.24)



Figure 4.24. Blue whiting otoliths from fish captured in the spring, during March.

4.6.2 Southern areas

• The first ring is usually well defined and rarely becomes dense. (Figure 4.25)



Figure 4.25. Blue whiting otoliths showing a well-defined first ring.

• The first ring can often be rather large compared to what is common in the otoliths from the northern areas. The inner ring can be over 12 mm in some cases. (Figure 4.26).



Figure 4.26. Blue whiting otoliths with an inner ring measuring 12.55mm.

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- The shape of the edge is used to distinguish between the youngest fish. 0-group fish have a "wavy" shape.
- When the wavy structure is not visible, and the first visible ring measures around 12mm, there are some steps to follow in order to establish the age (Figure 4.27):
 - Step 1: if the distance between the marked 1st and 2nd rings, is higher compared with the distance between the 2nd and the 3rd, even though the inner ring measurement is around 11-12mm, it should be considered as the true inner ring.
 - Step 2: look at the growth pattern between the marked 1st and 2nd rings, and the posterior rings and if the growth distance (1st 2nd rings) is narrow it could indicate a possible inner ring that is not visible. (Figure 4.27).



Figure 4.27. Blue whiting otoliths with the first visible ring measuring around 12mm.

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In this case the age classifications are made according with the following options:

• Option 1- Classified this otolith as an AQ3 – meaning that the reader does not trust the age classifications, due to the fact that the otolith is unreadable or very difficult to age with acceptable precision (Figure 4.28).



Figure 4.28. Blue whiting otoliths classified as AQ3, due to be very difficult to age with the acceptable precision.

- Option 2 Consider the possibility of having an inner ring that is not visible, and conduct the polishment technique on the otolith to clarify the existence of a not visible inner ring.
- Option 3 Confirm the first ring measurements for 0 and 1 age groups, in order to check if the first ring diameter fits into the range of measurements from the otoliths from the same area and quarter. If it fits it should be considered as the true first ring. If it doesn't fit, this possible indicates the presence of a not visible inner ring (faint ring).
- Step 3: if the ring distance between the marked/well defined 1st and 2nd rings, is higher compared with the distance between the 2nd and the 3rd, although the inner ring measurement is around 11-12mm, it should be considered as the true inner ring.

4.7 Otolith total length measurements

The blue whiting otoliths have been described as presenting a Bailey's zone (Bailey, 1970), a first mark which formation may be associated with a change of habit or depth; it would be equivalent to the "Bowers Zone" found in whiting otoliths (Gambell and Messtorff, 1964). In order to avoid considering this zone as the first annual ring, readers should measure the size of the inner ring, as described on blue whiting age reading guidelines (ICES WKARBLUE2 2017). The first annual ring is expected to present a diameter that ranges in size from 48 to 56 e.p.u. (corresponding to 8.00-9.33 mm) (ICES 2005). Although, a study conducted for the Portuguese coast (ICES division 27.9.a) showed that the diameter of first annual ring could change in size and reach a size as maximum as 12 mm wider (Dores and Gonçalves, 2017).

During the WKARBLUE3, one of the issues identified as an important source of uncertainty in age classifications is the position of the first annual ring. The otoliths included in the exchange were not enough to be used as conclusive in what concerns the first *annulus* size. Thus, the readers that participating in the workshop were asked to provide the total length of otoliths from 0 and 1 age groups. After the workshop, the participants provided the total otolith measurement by ICES area. The idea behind was that the total length of otoliths from ages 0 and 1, should give an indication of the expected range of the first annual ring by ICES area. The otoliths from age group 0 were from quarters 3 and 4 and otoliths from age group 1 were from quarters 1 and 2. This was decided because the peak spawning occurs during the first quarter, and it was therefore to be possible to follow the growth.

In these analysis only data from ages 0 and 1 have been used, the results revealed no significantly differences on the otoliths length distribution by sex (see details on the Working Document in Annex 7), because blue whiting only starts maturing at 2 years old (Bailey, 1982).

The otolith total length distributions (mm) by ICES area, quarter and age groups: age 0 (Figure 4.29) and age 1 (Figure 4.30) are presented on the following tables:

					ICE	S area			
		4a	5b	6a	7b	7g	7j	8c	9a
Age-group	Quarter	min-max	min-max	min-max	min-max	min-max	min-max	min-max	min-max
0	Q3	5.74 - 7.69						8.38 - 9.29	9.5 - 10.31
0	Q4		8.5 - 10	7.91 - 10.16	7.29 - 9.78	7.4 - 9.71	7.6 - 10.16	9.26 - 10.17	9.13 - 10.63

Figure 4.29	. Otoliths total l	ength in millimetres	(minimum and	maximum) and t	he number of	otoliths (n) b	y ICES area
and by qua	rter for the age g	group 0.					

		ICES area						
		2a	2a 5b 6a		8c	9a		
Age-group	Quarter	min-max (n)	min-max (n)	min-max (n)	min-max (n)	min-max (n)		
1	Q1			7.49 - 8.78 (30)	9.57 - 10.95 (30)	9.48 - 11.42 (55)		
1	Q2	8.53 - 10.7 (55)	9.4 - 11 (30)		9.61 - 11.13 (30)			

Figure 4.30. Otoliths total length in millimetres (minimum and maximum) and the number of otoliths (n) by ICES area and by quarter for the age group 1.

The measurements provided for each ICES area and quarter could be used as an indicator of the first ring measurement. Although, some concern and criticism should also be present on this application by the readers, mainly due to the reduced number of otoliths by ICES area, quarter and age group. In the future, more data should be added to allow to better define the first ring length distribution along the different ICES areas for blue whiting.

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5 Reference Collection

5.1 Northern areas

Otolith number	ICES area	Length (mm)	Sex	Age	Month	Comments
FO-68	5.b	210	Female	0	Nov	
lce-473634-81-9	5.a	200	Female	0	Nov	
FO-11	5.b	200	Female	1	Jan	
FO-15	5.b	200	Male	1	Jan	
Ice-473414-76-1	5.a	240	Male	1	Nov	
FO-06	5.b	250	Female	2	Jan	
Ice-473415-77-19	5.a	240	Female	2	Nov	
Noruega-23101-7	2.a	235	Female	2	May	
FO-14	5.b	260	Male	3	Jan	
FO-16	5.b	230	Male	3	Jan	
Ice-4676945-60-1	5.a	280	Female	3	July	
FO-07	5.b	250	Female	4	Jan	showing split ring before the 2nd ring
Ice-467499-41-18	5.a	250	Male	4	June	comment on edge!
FO-54	2.a	270	Male	4	Aug	
FO-09	5.b	300	Female	5	Jan	also showing split ring between 1 and 2
Ice-466265-24-11	5.a	270	Male	5	Mar	
Ice-473413-75-14	5.a	310	Male	5	Oct	
Ice-466134-8-25	5.a	310	Female	6	Mar	
NDL-5400110-98	6	301	Female	6	Mar	
Noruega-37687-3	2.a	320	Female	7	Aug	
Ice-466265-24-22	5.a	320	Male	8	March	AQ2 best example of age 8 could find
Ice-466270-29-9	5.a	360	Female	9	March	
Ice-466269-28-9	5.a	440	Female	12	May	

Figure 5.1. Otoliths selected for the reference collection from the Northern ICES areas.

The images of the otoliths from the reference collection from the Northern ICES areas are available in Annex 7.

Otolith number	ICES area	Length (mm)	Sex	Age	Month	Comments
IEO-Vigo-23	8.c	115	Undetermined	0	Nov	
IEO-Vigo-24	8.c	126	Undetermined	0	Oct	
Ipma-64	9.a	184	Female	0	Oct	
IEO-Vigo-29	8.c	218	Male	1	Nov	
IEO-Vigo-54	8.c	231	Male	1	Sept	
Ipma-3	9.a	195	Female	1	Jan	
IEO-Vigo-78	8.c	246	Female	2	Oct	
Ipma-11	9.a	235	Female	2	Feb	
Ipma-28	9.a	204	Male	2	April	
Ipma-7	9.a	219	Female	2	Jan	
IEO-Vigo-56	8.c	220	Male	3	Sept	
IEO-Vigo-64	8.c	250	Male	3	Aug	
Ipma-18	9.a	280	Female	3	Feb	
Ipma-38	9.a	271	Female	3	May	
IEO-Vigo-88	8.c	265	Male	4	Nov	
IEO-Vigo-13	8.c	273	Male	4	Aug	
IRL-2	7.c	240	Male	4	Mar	
IEO-Vigo-42	8.c	302	Female	5	Jun	
IEO-Vigo-47	8.c	352	Female	5	Jun	
IEO-Vigo-67	8.c	311	Female	5	Aug	
Ipma-59	9.a	297	Male	5	Aug	
IEO-Vigo-40	8.c	348	Female	6	April	
Ipma-21	9.a	312	Male	6	Feb	
IRL-10	7.c	290	Female	6	Mar	Good example of faint rings 1 and 2 and need to measure
Ipma-87	9.a	368	Female	6	Nov	
IEO-Vigo-95	8.c	371	Female	7	Oct	
IEO-Vigo-69	8.c	300	Male	8	Aug	
IRL-22	7.c	410	Female	10	Mar	

5.2 Southern areas

Figure 5.2. Otoliths selected for the reference collection from the Southern ICES areas.

The images of the otoliths from the reference collection from the Southern ICES areas are available in Annex 8.

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6 3-Point scale

Blue whiting age readings **should avoid otoliths classified as unreadable** or very difficult to interpret (0-25% reliability, classified as AQ3) according to the following 3-point scale of age reading quality that WKNARC (ICES. 2011) recommends to be used by all age readers who provide age data for stock assessments:

AQ1: Easy to age with high precision.

If a scale of 1-100 is applied, where 100 is when the reader has the highest possible confidence in the age reading and 1 is when the reader has no confidence in the age reading, age quality 1 (AQ1), will apply to approximately the top 25% of the possible quality ratings (75 – 100%). AQ1 is an indication that the age data is considered reliable for stock assessment.

AQ2: Difficult to age with acceptable precision.

Age quality 2 (AQ2), will apply approximately to age readings within 25 and 75 percentiles of the possible quality ratings. AQ2 is an indication that the age data is sufficiently reliable to be used for stock assessment purposes but improvement is required.

AQ3: Unreadable or very difficult to age with acceptable precision.

Age quality 3 (AQ3), will apply to approximately the lowest 25% of the possible quality ratings (0-25%). AQ3 is an indication that there are serious concerns about the reliability of the age data and/or its value to stock assessment WGs.

The blue whiting age readers are already following this procedure and the otoliths classified as AQ3 are not included on the data to be used for this species stock assessment (e.g. age length keys).

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7 Acknowledgements

We would like to thank to the main institutes that provided the otolith samples included on the exchange: Faroe Marine Research Institute (Faroe Islands), Instituto Español de Oceanografia (IEO, Spain), Institute of Marine Research (IMR, Norway), Marine Institute (MI, Ireland), Marine Laboratory Scotland (MLS, UK), Portuguese Institute for Sea and Atmosphere (IPMA, Portugal), Wageningen Marine Research (IMARES, The Netherlands).

We also thank to Dina Silva and Maria João Ferreira from IPMA (Portugal) for taking the otolith pictures that were used for the blue whiting age reading exchange in 2020 and also during the current workshop.

8 In memoriam

This report is dedicated to my lovely parents, Maria José J. V. Gonçalves (25/06/1951 to 11/11/2020) and Carlos Alberto P. Gonçalves (8/11/1947 to 26/12/2021), who are now bright stars in the sky.

With love,

From their daughter,

Patrícia Gonçalves
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Annex 2: Resolutions

The **Workshop on Age Reading of Blue Whiting** (*Micromesistius poutassou*) (WKARBLUE3), chaired by Patrícia Gonçalves, Portugal and Jane A. Godiksen, Norway met online, from May 31st to June 4th 2021 to address the following terms of reference:

- a) Review new information from validation study on first annual ring identification from daily increments; ICES Science plan 3.3, 5.1
- b) Review otolith growth table made by IPMA after WKARBLUE2 for aging of Blue whiting; ICES Science plan 3.3, 4.1
- c) Clarify the interpretation of annual growth rings (1-3) by sex, quarter and age through image analysis (measurements of ring distances and back calculation); ICES Science plan 3.1, 3.3, 4.1, 5.1
- d) Update on guidelines and common ageing criteria. With emphasis on testing the scheme made by WKARBLUE1.
- e) Increase existing reference collections of otoliths and improve the existing data base of otolith images.
- f) Analyse the age reading quality from the exchange using the 3-point scale of the image (mentioned in WKNARC).
- g) Address the generic ToR's adopted for workshops on age calibration (see 'PGCCDBS Guidelines for Workshops on Age Calibration').

The ToR a) hasn't addressed due to COVID-19 constraints which had preclude the development and application of new validation studies based on daily growth increments

Supporting Information

Priority:	Age determination is an essential feature in fish stock assessment to estimate the rates of moralities and growth. In order to arrive at appropriate management advice ageing procedures must be reliable.
	Otolith processing methods and age reading methods might differ considerably between countries. Therefore, otolith exchanges should be carried out on a regular basis, and if serious problems exist age reading workshops should be organised to solve these problems.
Scientific justification and relation to action plan:	The aim of the workshop is to review the available information on age determination, and val- idation for blue whiting, to identify the present problems in age determination for this spe- cies, improve the accuracy and precision of age determinations and spread information of the methods and procedures used in different ageing laboratories.
	A number of samples of otoliths (images) should be made available by the different laborato- ries to create a SamrtDots Event to assess the precision of age readers during 2024. Before the workshop, results from the otoliths circulation/exchange will be presented in 2025. Based on the exchange results, in 2025, age validation studies will be established to be conducted by the participants until the workshop. At the workshop, in 2025, results from the exchange and from the age validation studies will be presented and discussed.
Resource require- ments:	No specific resource requirements beyond the need for members to prepare for and participate in theexchange and in the meeting.
Participants:	In view of its relevance to the EU Data Collection Framework (DCF), the Workshop is expected to attract interest from ICES Member States.

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Secretariat facilities:	None.
Financial:	Additional funding will be required to facilitate the attendance of the scientists and techni- cians.
Linkages to advisory committees:	АСОМ
Linkages to other com- mittees or groups:	WGWIDE,WGBIOP, ACOM, RCMs, all WKACs (Age Calibration Workshops)
Linkages to other or- ganisations:	There is a direct link with the EU Data Collection Framework

Annex 3: Agenda

Monday 31 st	
09:00 - 09:30	Welcome and ToRs (Patrícia Gonçalves and Jane Godiksen)
09:30 - 10:00	Presentation of the results of the pre-workshop exchange (Jane Godiksen and Patrícia Gonçalves)
10:00 - 10:30	Presentation of issues regarding age readings and age reading protocol (ToR c and d) (Patrícia Gonçalves)
10:30 – 11:00	Presentation of validation study by IPMA (ToR b and c) (Patrícia Gonçalves)
11:00 – 11:30	Break
11:30 – 13:00	Go through pre-workshop exchange otoliths in SmartDots
13:00 – 13:30	Ending the day
Tuesday 1 st	
09:00 – 09:30	Presentation of the first ring issues and possible implications (Patrícia Gonçalves)
09:30 - 11:00	Go through pre-workshop exchange otoliths in SmartDots (continued)
11:00 – 11:30	Break
11:30 – 13:15	Go through pre-workshop exchange otoliths in SmartDots (continued)
13:15 – 13:30	Ending the day
<u> </u>	
weanesday 2 nd	
09:00 - 10:00	Update guidelines for age blue whiting in report Improve the age read- ing protocols (ToR d). Work in subgroups and report in plenary.
10:00 - 11:00	Work independently on new exchange using updated guidelines
11:00 – 11:30	Break
11:30 – 15:00	Work independently on new exchange using updated guidelines (con- tinued)

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Thursday 3 th	
09:00 - 11:00	Presentation of the results of yesterday's exchange (Patrícia Gonçalves)
11:00 – 11:30	Break
11:30 – 12:00 12:00 – 13:00	Review the guidelines based on the new exchange results - <i>working in subgroups (divide into north areas and south areas guidelines?)</i>
13:00 – 13:30	Review the guidelines based on the new exchange results – <i>subgroup leader reports to the plenary and plenary discussion</i>
	End of day, finish with an afternoon drink (bring your own)
Friday 4 th	
09:00 - 10:00	Create a reference collection of agreed age otoliths and make a training collection from each stock areas (ToR e)
10:00 - 11:00	Update guidelines for age blue whiting in report Improve the age reading protocols (ToR d)
11:00 – 11:30	Break
11:30 – 13:00	Update guidelines for age blue whiting (continued)
13:00 – 13:30	Sum up and goodbye

Annex 4: Contributions to the Workshop. Presentations and Working Documents.

Presentations: During the workshop a total of 6 presentations were performed, they are available and can be downloaded from the ICES WKARBLUE3 sharepoint.

The list of presentations is the following:

Patrícia Gonçalves and Jane Godiksen. Results from the pre-workshop exchange (see details in Section 3).

Patrícia Gonçalves. Blue whiting age readings – data and issues (see details Section 4).

Patrícia Gonçalves and Vítor V. Silva. Validation study on blue whiting age growth – IPMA table (part 2) (see details Section 2.2 and Annex 5).

Patrícia Gonçalves. Blue whiting age readings -first ring issues (see details Section 4).

Patrícia Gonçalves. Results from the workshop exchange WKARBLUE3 (see details Section 4.3).

Working Documents (see Annex 6):

Gonçalves, Patrícia; Ferreira, Ana Luísa; Diaz, Justine; Mullis, Eugene; Rasmussen, Helle; Coad Davies, Julia; Rodríguez, Lorena; Hernández, Carmen; Wilhelms, Ines; Kloppmann, Matthias; Godiksen, Jane Aanestad. Otolith total length measurements as a proxy to the inner ring size for blue whiting ageing.

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Annex 5: IPMA table (part 2)

ICES WKARBLUE3 - 31st May - 4th June 2021



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ICES WKARBLUE3 – 31st May – 4th June 2021

IPMA table (Part 2)

How to use: 2.Step

		Ring len	gth (mm)	Fish length (mm)			
	age	min	max	min	max	Stock_component	Sex
	0	9.47	10.75	164.11	200.47	Northern	Female
	1	10.75	11.86	200.47	231.59	Northern	Female
	2	11.86	12.95	231.59	260.96	Northern	Female
	3	12.95	14.01	260.96	288.70	Northern	Female
	4	14.01	15.04	288.70	314.88	Northern	Female
	5	15.04	16.05	314.88	339.59	Northern	Female
	6	16.05	17.02	339.59	362.93	Northern	Female
	7	17.02	17.98	362.93	384.96	Northern	Female
	8	17.98	18.91	384.96	405.75	Northern	Female
	0	10.16	11.12	172.01	203.82	Northern	Male
	1	11.12	11.99	203.82	229.92	Northern	Male
	2	11.99	12.86	229.92	253.60	Northern	Male
	3	12.86	13.73	253.60	275.09	Northern	Male
	4	13.73	14.59	275.09	294.58	Northern	Male
	5	14.59	15.46	294.58	312.26	Northern	Male
- 1	6	15.46	16.32	312.26	312.26 328.30 Northern		Male
	7	16.32	17.18	328.30	342.85	Northern	Male
	8	17.18	18.04	342.85	356.05	Northern	Male

ICES_Area: 27.5.a

Sex: M Otolith total length: 15.15 mm

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ICES WKARBLUE3 – 31st May – 4th June 2021





PMA Português do Mar e da Atmosfera

ICES WKARBLUE3 – 31st May – 4th June 2021

IPMA table (Part 2)

How to use: 4.Step

	Ring len	gth (mm)	Fish leng	th (mm)			
age	min	max	min	max	Stock_component	Sex	
0	9.47	10.75	164.11	200.47	Northern	Female	
1	10.75	11.86	200.47	231.59	Northern	Female	Catch date: 20-10-2017
2	11.86	12.95	231.59	260.96	Northern	Female	Edge: opaque
3	12.95	14.01	260.96	288.70	Northern	Female	Age: E
4	14.01	15.04	288.70	314.88	Northern	Female	Age: 5
5	15.04	16.05	314.88	339.59	Northern	Female]
6	16.05	17.02	339.59	362.93	Northern	Female	1
7	17.02	17.98	362.93	384.96	Northern	Female	1
8	17.98	18.91	384.96	405.75	Northern	Female	1
0	10.16	11.12	172.01		_		•
1	11.12	11.99	203.82	Adults	1		Immature (1-3 year olds)
2	11.99	12.86	229.92		Birth Date		Birth Date
3	12.86	13.73	253.60		Translucent	Opaque	ranslucent Gruppe Translucent
4	13.73	14.59	275.09	Mar			
5	14.59	15.46	294.58	Mor	thrank $\begin{bmatrix} 3 & F & M & X & M \\ 1 & 2 & 3 & 4 & 5 \end{bmatrix}$	6 7 8 9 10	$\begin{array}{c} 0 \\ 11 \\ 12 \\ \end{array} \begin{array}{c} \text{Monthrank} \\ 1 \\ 2 \\ 3 \\ 4 \\ 5 \\ 6 \\ 7 \\ 8 \\ 9 \\ 10 \\ 11 \\ 12 \\ \end{array}$
6	15.46	16.32	312.26		Teersteentuite	Tunt	Therefore a first star
7	16.32	17.18	328.30		Age = N	Age = N-1	Age = N Age = N-1
8	17.18	18.04	342.85				
					Opaque edge Age = N+1	Opaque edge Age = N	Opaque edge Age = N +1

IPMA Português do Mar e da do Mar e da

ICES WKARBLUE3 – 31st May – 4th June 2021

IPMA table (Part 2) How to use: 4.Step Ring length (mm) Fish length (mm) Catch date: 20-10-2017 Stock_component Sex age nin nax max 10.7 164.11 200.47 Northern Female 9.4 Edge: opaque 231.59 Northern 260.96 Northern 288.70 Northern 10.75 11.8 200.47 Female 1 Age: 5 11.86 12.9 231.59 Female Modal age: 5 (29% PA) (Pre-wk Exchange 2020) 12.95 14.01 3 260.96 Female 14.01 15.0 288.70 314.88 Northern Female Readers age classifications: 3-7 339.59 Northern 362.93 Northern 384.96 Northern 15.04 16.05 314.88 Female 5 16.05 17.02 17.02 17.98 339.59 Female 362.93 Female 17.9 18.9 384.96 405.75 Northern Female 0 10.16 11.12 172.01 Adults nature (1-3 year olds Im 11.99 12.86 11.12 203.82 Birth Dat Birth Date 11.99 229.92 2 12.86 13.7 253.60 13.73 14.59 275.09 J F M A M J J A S O N D 1 2 3 4 5 6 7 8 9 10 11 12 J F M A M J J A S O N D 1 2 3 4 5 6 7 8 9 10 11 12 4 Month Month Month Month 14.59 15.46 15.46 16.32 294.58 312.26 17.18 18.04 16.32 328.30 17.18 342.85 8 Opaque Age = N Ap

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IPMA table (Part 2) - indicator of possible false or double rings





Age reader classification ≠ 5

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IPMA table (Part 2) - How to use: 5. Step - check possible false or double rings





Age reader classification ≠ 5

Double check the age rings: By looking at the entire structure of the otolith and following the sequence of yearly growth



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IPMA table (Part 2) -How to use: 5. Step - check possible false or double rings





Age reader classification ≠ 5



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IPMA table (Part 2) - How to use: 6. Step - Age classification



After steps: 1-5

Age classification \mathbf{V}



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IPMA table (Part 2)

Southern

	Ring len	gth (mm)	Fish leng	gth (mm)		
age	min	max	min	max	Stock_component	Sex
0	9.62	11.09	175.63	212.23	Southern	Female
1	11.09	12.38	212.23	243.64	Southern	Female
2	12.38	13.63	243.64	273.36	Southern	Female
3	13.63	14.84	273.36	301.49	Southern	Female
4	14.84	16.01	301.49	328.11	Southern	Female
5	16.01	17.15	328.11	353.30	Southern	Female
6	17.15	18.25	353.30	377.14	Southern	Female
7	18.25	19.33	377.14	399.71	Southern	Female
8	19.33	20.27	399.71	418.98	Southern	Female
0	9.62	11.09	177.04	212.16	Southern	Male
1	11.09	12.38	212.16	239.01	Southern	Male
2	12.38	13.63	239.01	261.78	Southern	Male
3	13.63	14.84	261.78	281.09	Southern	Male
4	14.84	16.01	281.09	297.47	Southern	Male
5	16.01	17.15	297.47	311.35	Southern	Male
6	17.15	18.25	311.35	323.13	Southern	Male
7	18.25	19.33	323.13	333.12	Southern	Male
8	10.22	20.27	222 12	3/10 80	Southern	Male

Northern

	Ring length (mm) Fish length (n					
age	min	max	min	max	Stock_component	Sex
0	9.47	10.75	164.11	200.47	Northern	Female
1	10.75	11.86	200.47	231.59	Northern	Female
2	11.86	12.95	231.59	260.96	Northern	Female
3	12.95	14.01	260.96	288.70	Northern	Female
4	14.01	15.04	288.70	314.88	Northern	Female
5	15.04	16.05	314.88	339.59	Northern	Female
6	16.05	17.02	339.59	362.93	Northern	Female
7	17.02	17.98	362.93	384.96	Northern	Female
8	17.98	18.91	384.96	405.75	Northern	Female
0	10.16	11.12	172.01	203.82	Northern	Male
1	11.12	11.99	203.82	229.92	Northern	Male
2	11.99	12.86	229.92	253.60	Northern	Male
3	12.86	13.73	253.60	275.09	Northern	Male
4	13.73	14.59	275.09	294.58	Northern	Male
5	14.59	15.46	294.58	312.26	Northern	Male
6	15.46	16.32	312.26	328.30	Northern	Male
7	16.32	17.18	328.30	342.85	Northern	Male
8	17.18	18.04	342.85	356.05	Northern	Male

Annex 6: Working Documents

Otolith total length measurements as a proxy to the inner ring size for blue whiting ageing

Gonçalves, Patrícia¹; Ferreira, Ana Luísa¹; Diaz, Justine²; Mullis, Eugene³; Rasmussen, Helle⁴; Coad Davies, Julia⁴; Rodríguez, Lorena⁵; Hernández, Carmen⁵; Wilhelms, Ines⁶; Kloppmann, Matthias⁶; Godiksen, Jane Aanestad²

¹ IPMA(Portugal); ² IMR (Norway); ³ MI (Ireland); ⁴ DTU-Aqua (Denmark); ⁵ IEO (Spain); ⁶ TI (Germany).

The blue whiting otoliths have been described as presenting a Bailey's zone (Bailey, 1970), a first mark which formation may be associated with a change of habit or depth; it would be equivalent to the "Bowers Zone" found in whiting otoliths (Gambell and Messtorff, 1964). In order to avoid considering this zone as the first annual ring, readers should measure the size of the inner ring, as described on blue whiting age reading guidelines (ICES WKARBLUE2 2017). The first annual ring is expected to present a diameter that ranges in size from 48 to 56 e.p.u. (corresponding to 8.00-9.33 mm) (ICES 2005). Although, a study conducted for the Portuguese coast (ICES division 27.9.a) showed that the diameter of first annual ring could change in size and reach a size as maximum as 12 mm wider (Dores and Gonçalves, 2017).

During the WKARBLUE3, one of the issues identified as an important source of uncertainty in age classifications is the position of the first annual ring. The otoliths included in the exchange was not enough to be used as conclusive in what concerns the first *annulus* size. Thus, the readers that participating in the workshop were asked to provide the total length of otoliths from 0 and 1 age groups. After the workshop, the participants provided the total otolith measurement by ICES area. The main goal is using the total length of otoliths from ages 0 and 1, to have an indication of the expected range of the first annual ring size by ICES area. The otoliths from age group 0 were from quarters 3 and 4 and otoliths from age group 1 were from quarters 1 and 2. This is because the peak spawning occurs during the first quarter, and this way it was possible to follow the growth pattern.

Data

After the workshop, participants provided the measurements from the otoliths total length of age groups 0 and 1, as shown in Figure A1a and A1b, respectively.

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Figure A1. Total length of the otoliths (mm) for age group 0 (a) and age group 1 (b).

A summary table (Table 1) shows the number of measurements provided from each age group and each ICES area, by each of the Member States (MS).

Table A1. The number of otoliths from 0- and 1-age group by ICES area and provided by each Country (MS) is described below:

		ICES area									
Country (MS)	27.2.a	27.4.a	27.5.b	27.6.a	27.7.b	27.7.g	27.7.j	27.8.c	27.9.a	Total	
Germany											
0 -age group			30							30	
1-age group	25	30								55	
Denmark											
1-age group	30									30	

Ireland										
0 -age group				30	30	30	30			120
Norway										
0 -age group		25								25
1-age group				30						30
Portugal										
0 -age group									22	22
1-age group									55	55
Spain										
0 -age group								60		60
1-age group								60		60
Total	55	55	30	60	30	30	30	120	77	487

The number of otoliths measured by ICES area and by each sex is shown in Figure A2.



Figure A2. The total number of otoliths measured by ICES area and by sex (F- Female, M – Male, I – Undetermined).

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Results

The otoliths total length (mm) revealed differences between ICES areas, for the two age groups. Those differences are represented on the next figure (Figure A3):



Figure A3. Otolith total length (mm) by ICES area and by age groups, 0 and 1. The blue dashed line represents the 8 mm which, according to the literature, corresponds to the lower range limit for the first annual ring. The blue solid line represents 9.33 mm which, according to the literature, corresponds to the upper range limit for the first annual ring.



Figure A4. Otolith total length (mm) by ICES area, by age groups (0 and 1) and by quarter. The blue dashed line represents the 8 mm which, according to the literature, corresponds to the lower range limit for the first annual ring. The blue solid line represents 9.33 mm which, according to the literature, corresponds to the upper range limit for the first annual ring.

The fish length distribution of the samples and the otoliths total length by ICES area and age group is summarized in tables A2 and A3, respectively.

Table A2. Fish total length in millimetres (minimum and maximum), by age group, by ICES area and by sex (F - Female, M – Male, I – Undetermined).

			Fish Total Length (min - max)											
Age group	Sex	27.2.a	27.4.a	27.5.b	27.6.a	27.7.b	27.7.g	27.7.j	27.8.c	27.9.a				
0	F		125 - 165	180 - 210	170 - 200	140 -210	150 - 210	140 - 200	171 - 211	170 - 205				
0	I			200 - 210	140 - 200	140 - 170	140 - 190							
0	м		110 - 155	170 - 210	140 - 200	140 - 200	150 - 190	140 - 190	180 - 220	170 - 210				
1	F	170 - 220	190 - 230		160 - 190				189 - 222	188 - 220				
1	м	170 - 230	190 - 230		160 - 195				192 - 236	175 - 220				
1	I	200 - 220	200 - 220											

Table A3. Otoliths total length in millimetres (minimum and maximum), by age group, by ICES area and by sex (F - Female, M – Male, I – Undetermined).

		Otolith Total Length (min - max)									
Age group	Sex	27.2.a	27.4.a	27.5.b	27.6.a	27.7.b	27.7.g	27.7.j	27.8.c	27.9.a	
0	F		6.19 - 7.69	9.2 - 10	8.73 - 9.65	7.29 - 9.78	7.64 - 9.71	7.89 - 10.16	8.38 - 9.95	9.13 - 10.31	
0	I			9.8 - 9.9	7.91 - 10.16	7.73 - 8.55	7.67 - 8.89				
0	м		5.74 - 7.32	8.5 - 9.9	8.03 - 9.4	7.72 - 9.28	7.4 - 9.28	7.6 - 8.84	8.5 - 10.17	9.42 - 10.63	
1	F	8.53 - 10.7	9.4 - 11		7.81 - 8.67				9.57 - 10.57	9.86 - 11.42	
1	м	8.79 - 10.2	9.4 - 10.5		7.49 - 8.76				9.59 - 11.13	9.48 - 11.42	
1	I	9.7 - 10.5	9.5 - 10.5								

The results revealed no significantly differences on the otoliths length distribution by sex (Table A3). Since all the individuals analysed where immature, because blue whiting only starts maturing at 2 years old (Bailey, 1982).

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Figure A5. Fish total length (mm) by otolith total length (mm) by each ICES area, by age groups (0 and 1) and by sex. The sex variable is represented by the symbols: circle (females), square (males) and triangle (undetermined).



Figure A6. Fish total length by otolith total length (mm) by age groups (0 and 1) and by each ICES area.

The next tables present the otolith total length distributions by ICES area, quarter and age groups: age 0 (Table A4) and age 1 (Table A5).

Table A4 – Otoliths total length in millimetres (minimum and maximum) by ICES area and by quarter for the age group 0.

		ICES area							
		4a	5b	6a	7b	7g	7j	8c	9a
Age-group	Quarter	min-max	min-max	min-max	min-max	min-max	min-max	min-max	min-max
0	Q3	5.74 - 7.69						8.38 - 9.29	9.5 - 10.31
0	Q4		8.5 - 10	7.91 - 10.16	7.29 - 9.78	7.4 - 9.71	7.6 - 10.16	9.26 - 10.17	9.13 - 10.63

Table A5 – Otoliths total length in millimetres (minimum and maximum) by ICES area and by quarter for the age group 1.

		ICES area							
		2a	5b	6a	8c	9a			
Age-group	Quarter	min-max	min-max	min-max	min-max	min-max			
1	Q1			7.49 - 8.78	9.57 - 10.95	9.48 - 11.42			
1	Q2	8.53 - 10.7	9.4 - 11		9.61 - 11.13				

References

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Gambell, R., Messtorff, J. 1964. Age determination in the whiting. Journal du Conseil International pour l'Exploration de la Mer. 28, 393-404.

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Annex 7: Northern areas – reference collection



Age 1



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Age 6



Age 7













Annex 8: Southern areas – reference collection



Age 1











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Age 5













Age 8



