ICES Cooperative Research Report

Rapport des Recherches Collectives

No. 294

April 2009

Hake age estimation: state of the art and progress towards a solution

Editors

Carmen G. Piñeiro • Cristina Morgado

Maria Saínza • William James McCurdy

ICES
International Council for the Exploration of the Sea

CIEM
Conseil International pour l'Exploration de la Mer

International Council for the Exploration of the Sea
Conseil International pour l'Exploration de la Mer

H. C. Andersens Boulevard 44–46
DK-1553 Copenhagen V
Denmark
Telephone (+45) 33 38 67 00
Telefax (+45) 33 93 42 15
www.ices.dk
info@ices.dk

Recommended format for purposes of citation:

Piñeiro, C. G., Morgado, C., Saínza, M., McCurdy, W. J. (Eds). 2009. Hake age estimation: state of the art and progress towards a solution. ICES Cooperative Research Report No. 294. 43 pp.

ISBN: 87-7482-004-4

ISSN: 1017-6195

Contents

Executive summary 1

1 Introduction 2

2 Review of otolith age estimation calibration exercises: exchanges and workshops 4

2.1 Overview of otolith age-reading exchanges and workshops 4

2.2 Summary of the most recent (2004) hake otolith workshop report 11

2.2.1 Introduction 11

2.2.2 Material and methods 11

2.2.3 Results and discussion 13

2.2.4 Conclusions 14

2.2.5 Recommendations 14

3 Summary of current knowledge 16

3.1 Age-reading criteria 16

3.2 Reproductive behaviour 18

3.3 Length–frequency analysis 20

3.4 Growth parameters 20

3.5 Bioenergetics studies 22

3.6 Daily growth 22

3.7 Mark and recapture 24

4 Key issues 28

4.1 Complexity of the otolith growth pattern 28

4.2 Lack of validated ageing criteria 29

4.3 Alternative methods 30

4.4 Fast growth vs. slow growth 31

5 Future work 32

5.1 Exchange programme 32

5.2 Otolith biometrics 32

5.3 Otolith macrostructure typology 32

5.4 Experiments in a controlled environment 33

5.5 Developing a new protocol for hake age estimation 33

5.6 Summary of recommendations for future work 33

Acknowledgements 34

6 References 35

Acronyms 42

Executive summary

Since 1992, northern and southern hake (*Merluccius merluccius*) stock assessments have used age data based on otolith analysis. Age data for stock assessment is provided by different institutions, which implies a quantification of age-reading precision to estimate assessment quality indicators. During this period, considerable effort has been made to improve the precision of age data by means of successive age-reading calibration exercises, exchanges, and workshops in 1997, 1999, 2001, and 2004. This goal was partly achieved, and experts recently agreed on standard criteria (Piñeiro and Saínza, 2003) that allowed an acceptable precision to be reached for ages up to 3 years (Piñeiro *et al.*, 2004). However, these criteria have never been validated, and recent mark–recapture experiments are not in line with ageing results based on the standard criteria.

Given the impact of bias in age estimation on stock assessment results, consequent management advice, and concern about the state of the hake stocks (ICES, 2007a, 2007b), a report on the current state of the art is needed. The main goal of this report is to present a synthesis of the work carried out over the years by researchers involved in providing age data for stock assessment, mainly on age-reading calibration exercises, and current knowledge regarding the growth and ageing of this species. This report also includes recommendations for future work aimed at achieving validated age-reading criteria.

1 Introduction

The European hake (*Merluccius merluccius* Linnaeus, 1758) is one of the most valuable and heavily exploited demersal species in the western European fisheries. It is widely distributed in the Northeast Atlantic, from Norway to Mauritania, being more abundant from the British Isles to south of Spain (Casey and Pereiro, 1995) and in the Mediterranean and Black seas. It is found mainly at depths between 50 and 370 m, although its distributional bathymetric range is 30–1000 m (Domínguez-Petit, 2007). Hake is a batch-spawner (Murúa *et al.*, 1996; Domínguez-Petit, 2007) with a protracted spawning season extending throughout the entire year, but mainly from December to July along the continental shelf edge.

The assessment of hake is undertaken annually by the ICES Working Group on the Assessment of Southern Shelf Stocks of Hake, Megrim, and Monkfish (WGHMM). Since 1978, despite the lack of a sound biological basis, ICES has distinguished two separate hake stocks for management purposes: (i) the northern stock (ICES Division IIIa, Subareas IV, VI, and VII, and Divisions VIIIa–b); and (ii) the southern stock (ICES Divisions VIIIc and IXa), split at Cape Breton Canyon (ICES, 1979). Total annual landings of European hake declined from ca. 125 000 t in the early 1960s to the current level of ca. 44 000 t for the northern stock and 15 000 t for the southern stock. In recent years, the relatively depleted levels of both stocks have been a cause for concern to ICES (ICES, 2007a, 2007b).

The assessments for hake include several areas of uncertainty, such as growth, population structure, and stock definition, with growth being one of the most limiting factors for stock assessment. Currently, both stock assessments use age-structured models. Age data are provided by different countries and follow agreed international ageing criteria (Piñeiro and Saínza, 2003) based on the results of successive international age-reading exercises and workshops (held in 1997, 1999, 2001, and 2004). A working document presented at the 2002 WGHMM meeting demonstrated that the northern hake assessment had a better fit to the assessment model if the plus group were set at age 10 instead of age 8 (ICES, 2003). However, the confident ages from the previous otolith age estimation workshop ranged from 0 to 5 years and, because the scientists involved in hake ageing and growth drew the attention of WGHMM to the low reliability of age 10, the working group decided to continue to use age 8 as the plus group.

The ages recorded for both stocks of hake usually range from 0 to 12 years, although the landing and survey data are dominated by specimens belonging to age groups up to three years.

The assessment of the southern hake stock has been based on age-reading data since the 1999 assessment, with age–length keys (ALKs) from 1992 onwards (ICES, 2000). The northern stock assessment used ALKs based on otolith readings from 1992 (ICES, 1994a). The ALKs that were applied to catch data prior to 1992 are described in the WGHMM report (ICES, 2007a). For both stocks, when the assessment was performed without ALKs, the age composition of the catches was estimated using a numerical method developed by Kimura and Chikuni (1987).

In hake, the *sagitta* otoliths are routinely used for ageing. The annual rings (*annuli*) have proven difficult to interpret because of the complexity of the otolith macrostructure, and the age estimation method currently in use has not been validated. Recently, however, progress has been made regarding the precision of age data. More than five exchanges and four workshops have taken place in the past two decades. The lack of high precision, despite these efforts, is a strong indication of the difficulty of hake age interpretation.

At the end of 1990s, within the framework of EU Study Contract 95/038 "Biological Studies of Demersal Fish" (BIOSDEF; see Anon., 1998), standard criteria for the ageing of hake otoliths were developed. These criteria were adopted by age readers from all the institutions involved in hake stock assessment.

In recent years, new relevant knowledge has been obtained concerning the growth of this species, based on the mark–recapture method (de Pontual *et al.*, 2003; Piñeiro *et al.*, 2007). Results based on a blind interpretation of marked hake otoliths concluded that age estimates were neither accurate nor precise and invalidated the age estimation method (de Pontual *et al.*, 2006). The results from mark–recaptures are not compatible with the growth rate obtained by mean otolith age estimation, which raises concerns about the use of potentially inaccurate age data for stock assessment and resulting management advice. The impact of biased age estimates on European hake stock assessment is currently under investigation (Bertignac and de Pontual, 2007).

The main goal of this report is to summarize the current status of age estimation for European hake and the work carried out over the years by researchers involved in providing age data for stock assessment purposes. The current knowledge of the age and growth of this species will be described, together with the results of recent international exchanges and workshops held before and after the availability of hake growth information from mark–recapture programmes. In addition, guidelines are provided with a view to furthering progress in ageing studies of this species. Considering the long history of efforts devoted to resolving this problem and the need to have reliable age-structure data for stock assessment, this review is long overdue.

2 Review of otolith age estimation calibration exercises: exchanges and workshops

2.1 Overview of otolith age-reading exchanges and workshops

Although several hake ageing problems have yet to be addressed, several advances have taken place in the past two decades that have involved many scientists from different countries. Table 2.1.1 summarizes the main exchanges and workshops carried out to date. It should be mentioned that several researchers and otolith readers have devoted special attention to the improvement of age-reading precision and the establishment of an internationally agreed ageing protocol for this species. The revision of the previous calibration exercises is important because the outcomes and reports from these exercises have not been published and are collected in contributions to ICES working groups and project reports. The ICES Planning Group on Commercial Catch, Discards, and Biological Sampling (PGCCDBS) provides access to reports on age calibration (age reading) exchanges and workshops via its document repository at http://www.ices.dk/reports/acfm/pgccdbs/PGCCDBSdocrepository.asp.

In the 1980s, the scientists involved in hake age reading for stock assessment purposes also used the data to determine growth parameters, whose wide range suggests that different age-reading methods were employed. To detect these differences, several international *ad hoc* working groups and exchanges were conducted (FAO, 1982; ICES, 1983, 1984, 1986; Oliver *et al.*, 1989). The analysis performed was based mainly on percentage of agreement; the low precision of the results indicated that age interpretation of hake otoliths was very complex and further work was needed on standardization of protocols (Table 2.1.1).

A workshop preceded by an exchange was conducted in 1994 between IFREMER (French Research Institute for the Exploitation of the Sea) and IEO (Spanish Institute of Oceanography), based on the recommendations of the ICES Workshop on Sampling Strategies for Age and Maturity (ICES, 1994b). The main objective of this exchange was to assess age-reading precision, using the recommended protocol (Piñeiro and Meixide, 1995). At the workshop, the general criteria adopted were based on the number of annual rings (consisting of one opaque and one translucent, or hyaline, zone) and used 1 January as the birthday. Readers were not made aware of the length of the fish. The readings were carried out using a stereomicroscope (magnification × 20) under reflected light, but transmitted light was also used to help discriminate between translucent and opaque growth rings. A video camera connected to a large screen was used to facilitate discussion of the interpretation criteria. The results indicated a low level of agreement between the two readers. Since that time, the guidelines for organizing exchanges and workshops, the methods used, and the analysis of the data have remained unchanged.

Within the framework of BIOSDEF (Anon., 1998), a workshop was organized in 1997 (Piñeiro, 2000) to improve the method of ageing hake in countries that share the same stocks. Prior to the workshop, an exchange was carried out during 1995/1996. The otolith readers applied the protocol used at the 1994 workshop, and the analysis of the age-reading results was carried out using the methodology proposed by Eltink (1997). Following the recommendation given by ICES (1994), several statistical indices and tests were used for analysing the results:

- Wilcoxon's rank test, to assess bias between a pair of readers
- bias plots of the average age, ±2 s.d. (standard deviation), obtained by each age reader; all age reader results were plotted against modal age, considered as the referential age
- average percentage error (APE), from Beamish and Fournier (1981), to assess a measure of the precision of a series of readings
- mean coefficient of variation (CV)
- index of precision (Chang, 1982)
- box-whisker plots for visual presentation of the observations

These analyses provided information on the precision of age readings by age group for both individual readers and all readers combined. An interpretation criterion for the first three ages was agreed, but precision levels were low for the age-reading exercises performed. However, this workshop was considered to be a further step towards reaching a consensus for age-reading criteria (Table 2.1.1). It was agreed that more training was necessary to increase precision levels, and a further exchange was recommended.

Another workshop was carried out in 1999 to analyse results from the exchange circulated in the previous year (Piñeiro *et al*., 2000). This workshop was planned within the framework of EU Study Contract No. 97/015 "New Assessment and Biology of the Main Commercial Fish Species: Hake and Anglerfish of Southern Shelf Demersal Stocks in the South Western Europe" (DEMASSESS; see Anon., 2000). New readers started to be involved in hake otolith ageing, and their participation at the workshop was considered worthwhile in facilitating their understanding of the ageing criteria. Two otolith samples prepared by the IFREMER and IEO laboratories were read during the exchange, and a copy of the protocol used at the last workshop (1997) was provided to all participants. Fish length information from IEO otolith collections was available and digital images of sectioned otoliths were used and circulated for the first time. For the age-reading analysis, a more extensive study was performed to compare the age interpretations of the readers. However, the Wilcoxon's rank test was not used because it was considered inappropriate to perform multiple paired comparisons when more than two readers were involved in ageing the same collection (Zar, 1996). In order to determine which groups of readers showed the higher levels of agreement, cluster analysis and multiscaling dimensions were used.

For reproducibility measures (APE and CV), the values for all readers in general had decreased since the last exchange, particularly for the IFREMER sample. The CV of the expert readers was reduced, but the APE index remained the same (Figure 2.1.1). It should be emphasized that these measures of agreement should be interpreted with caution because of the influence of sample size and, in particular, of younger ages in CV calculations. At the 1999 workshop, it was recognized that the level of reader experience is an important aspect of the achievement of higher precision for hake age reading, particularly in the identification of false (non-annual) rings. Results indicated that it is possible to use the annual ALK instead of numerical methods to estimate age composition of catches for the southern hake stock assessment, taking into account the observed agreement between the readers involved.

Until this time, the age-structure composition of the northern hake stock catches had been based solely on IFREMER ALKs. However, in light of the agreement on age reading that was reached, the possibility of applying ALKs from AZTI-Tecnalia (the Basque Technological Centre for Marine and Food Investigation) in addition to those from IFREMER was recommended. The agreement between readers for ages 0–4 was improved compared with the previous workshop (1997), mainly in the identification of the first annual ring. Despite improvements in precision for all readers, the results continued to highlight the difficulties in interpreting the age of hake, and it was recommended to hold another exchange and workshop in two years. Furthermore, it was recommended that alternative techniques (tagging, microchemistry, etc.) for validation of the age estimation of this species be explored.

Under EU Contract No. 99/009 "Improving Sampling of Western and Southern European Atlantic Fisheries" (SAMFISH), an otolith exchange on hake was undertaken in 2001 (Piñeiro and Saínza, 2002) from both northern and southern ICES stocks in order to calibrate the age readings and to estimate the precision level. The terminology, guidelines, and tools used for age-reading comparison were carried out according to the recommendations of Eltink *et al.* (2000). Readers had access to the collection of otoliths used in the exchange and to the digitized images from the otolith sections. Information on catch date, sex, and total length were available to the readers. The same procedures as for the previous exchange were followed (1998), and the exploratory and statistical analyses revealed the highest level of agreement in the location of the first three annual rings as a result of the adoption of the ageing criteria. The results indicated that, although an improvement in precision was evident for almost all readers, the estimation of older fish (> 5 years) appeared to be the major difficulty. The latter result justified the need for another exchange among readers that provided stock assessment age data, with particular focus on higher ages.

In 2002, the ICES WGHMM demonstrated the difficulties in the assessment of hake that result from uncertainty in age estimation for older fish, which led the working group to use a plus group at age 8. To deal with these problems, an otolith exchange focusing mainly on older fish was conducted in 2003 between readers involved in the assessment (Piñeiro *et al.*, 2004). In contrast to the previous exchange (2001), no information on fish length was available to the readers. The exchange results indicated that the precision of age readings had decreased. The comparison of the results obtained from this exchange (2003) and the previous exchange (2001; Figure 2.1.1) highlights the difficulty of hake age estimation, and it was recommended that an international workshop specifically aimed at solving these problems should be held.

The 2003 exchange demonstrated a strong bias in age estimations of older fish. The precision (CV) of age estimation had decreased from ca. 17 to 32% for the ALK readers between the previous two exchanges (2001, 2003) for otolith collections within the same fish length range (Figure 2.1.1). The age range regarded by age readers as confident also dropped: from 0–5 years to 0–3 years. This negative result was a consequence of the difficulty of hake otolith reading using non-validated age determination criteria. The lack of knowledge of fish length in this exchange, in contrast to the availability of fish length information during the 2001 exchange, could also contribute to the reduction in precision and the "confident" age range. This mainly affected less experienced readers, who were not accustomed to measuring ring radius when they performed routine hake age readings.

An international age-reading workshop was conducted in 2004 to tackle the problem of providing age estimates of fish older than 5 years (see Section 2.2).

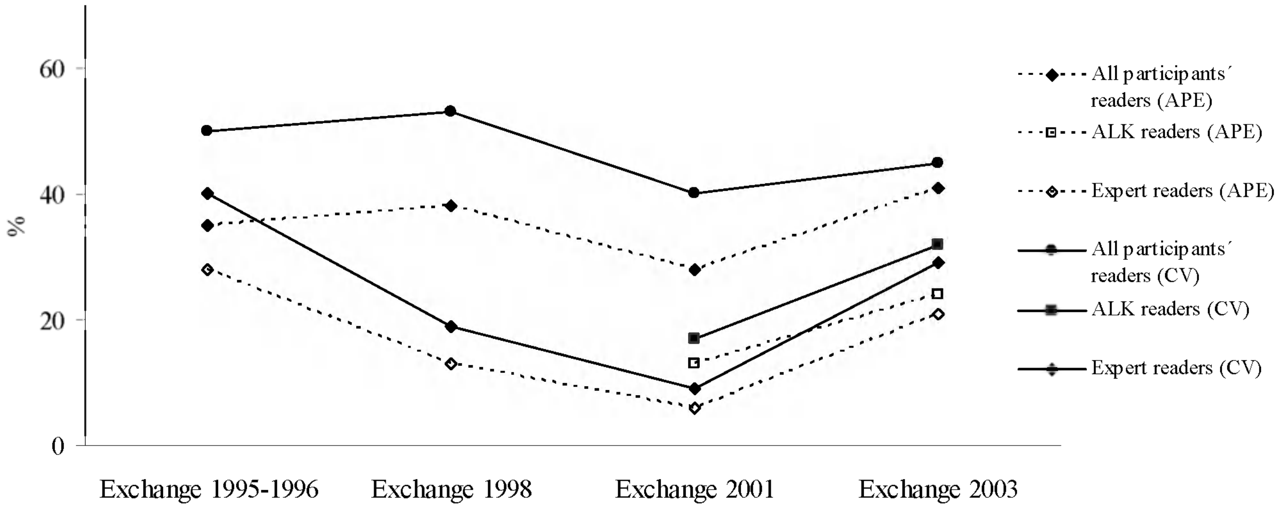

Figure 2.1.1. Average percentage error (APE) from Beamish and Fournier (1981) and coefficient of variation (CV) for exchanges between 1995/1996 and 2003 for all participants, readers that provided age data for stock assessment purposes (ALK readers), and expert readers.

Table 2.1.1. Overview of hake age estimation events conducted during the past three decades, with information on sample sets read, objectives, and achievements.

Year	Event	Participating countries	Area	No. of readers	Sample: no. otoliths & length range (TL cm)	Objectives	Main results
1981	Workshop methodologies used for fish age reading	France, Italy, Morocco, Senegal, and Spain	Mediterranean NE Atlantic	14		Compare preparation techniques and reading methods	No standard method agreed

1983	Workshop	France, Spain	NE Atlantic	4		Preparation techniques selection and agreed age-reading criteria	• Standard preparation technique adopted using transversal sections of otoliths
							• Improvement in agreement on otolith interpretation, but no standardization
							• Identification of three sources of different interpretation:
							  • difficulty in locating first *annulus*
							  • discrimination of true rings from checks
							  • interpretation of the edge

1984	*Ad hoc* WG	Canada, France, and Spain	NE Atlantic	5			
1986	*Ad hoc* WG	Canada, France, Portugal, Spain, and UK	NE Atlantic	14			
1989	Workshop	France, Greece, Spain, and Italy	Mediterranean	9			
1994	Exchange and workshop	France and Spain	NE Atlantic	2	95 (15 – 67)	Assess age-reading precision	• Main difficulties related to correct identification of first *annulus* • No difference in growth pattern between otoliths from northern and southern stocks Readers / Agreement between first and second reading / CV (%) (intra-reader): Spanish / 43 – 98 / 75 French / 43 – 98 / 50
1995 – 1996	Exchange	France, Portugal, Spain, and UK	NE Atlantic	7	192 (16 – 55)	Assess age-reading precision and agreed age-reading criteria	Readers / APE (%) / CV (%): All readers / 35 / 50 ALK readers / 28 / 39

<table>
<tr><th>Year</th><th>Event</th><th>Participating countries</th><th>Area</th><th>No. of readers</th><th>Sample: no. otoliths & length range (TL cm)</th><th>Objectives</th><th>Main results</th></tr>
<tr><td>1997</td><td>Workshop</td><td>France, Portugal, Spain, and UK</td><td>NE Atlantic</td><td>9</td><td>95
(16 – 55)</td><td>Assess age-reading precision and agreed age-reading criteria</td><td>

Otolith interpretation criteria agreed for first three ages
No difference in growth pattern between otoliths from both stocks
Reference factor adopted to locate first annulus
Two or three checks before first annulus
Difficulty interpreting otolith edge

<table>
<tr><th></th><th>APE (%)</th><th>CV (%)</th></tr>
<tr><td>All readers</td><td>16</td><td>21</td></tr>
<tr><td>ALK readers</td><td>11</td><td>16</td></tr>
</table>
</td></tr>
<tr><td>1998</td><td>Exchange</td><td>France, Portugal, Spain, and UK</td><td>NE Atlantic</td><td>11</td><td>100 (IEO)
(12 – 69)
107 (IFREMER)
(19 – 102)</td><td>Assess age-reading precision and agreed age-reading criteria</td><td>
<table>
<tr><th></th><th colspan="2">APE (%)</th><th colspan="2">CV (%)</th></tr>
<tr><td>Sample sets</td><td>IEO</td><td>IFREMER</td><td>IEO</td><td>IFREMER</td></tr>
<tr><td>All readers</td><td>37</td><td>33</td><td>53</td><td>44</td></tr>
<tr><td>ALK readers</td><td>13</td><td>5</td><td>19</td><td>7</td></tr>
</table>
</td></tr>
<tr><td>1999</td><td>Workshop</td><td>UK, France, Ireland, Portugal, and Spain</td><td>NE Atlantic</td><td>11</td><td>54 (IEO)
(12 – 69)
41 (IFREMER)
(20 – 76)</td><td>Assess age-reading precision and agreed age-reading criteria</td><td>

Adoption of ageing criteria for ages 0 – 4
High level of variability between readers for ages greater than 5
High agreement in locating first annulus
Precision level highly related to ageing experience
Difficulty interpreting age patterns on hake otoliths justifies need for periodic exchange exercises

<table>
<tr><th></th><th colspan="2">APE (%)</th><th colspan="2">CV (%)</th></tr>
<tr><td>Sample sets</td><td>IEO</td><td>IFREMER</td><td>IEO</td><td>IFREMER</td></tr>
<tr><td>All readers</td><td>29</td><td>21</td><td>42</td><td>31</td></tr>
<tr><td>ALK readers</td><td>13</td><td>10</td><td>13</td><td>14</td></tr>
</table>
</td></tr>
</table>

<table>
<tr><th>Year</th><th>Event</th><th>Participating countries</th><th>Area</th><th>No. of readers</th><th>Sample: no. otoliths & length range (TL cm)</th><th>Objectives</th><th>Main results</th></tr>
<tr><td>2001</td><td>Exchange</td><td>UK, France, Ireland, Portugal, and Spain</td><td>NE Atlantic</td><td>12</td><td>199
(11 – 83)</td><td>Assess age-reading precision and improvement in agreed age-reading criteria; new readers started to be in-volved in otolith ageing</td><td>

Highest level of agreement obtained so far in location of first three annuli as result of using estab-lished ageing criteria
Highest level of agreement (%) achieved for first 5 age groups
APE and CV indices for all readers are best from all exchanges
No significant differences between otoliths from both stocks (p > 0.05)

<table>
<tr><th>Readers</th><th>Agreement (%)</th><th>APE (%)</th><th>CV (%)</th></tr>
<tr><td>All</td><td>58</td><td>28</td><td>40</td></tr>
<tr><td>ALK</td><td>75</td><td>13</td><td>17</td></tr>
<tr><td>Expert</td><td>84</td><td>6</td><td>9</td></tr>
</table>
</td></tr>
<tr><td>2003</td><td>Exchange</td><td>UK, France, Ireland, Portugal, and Spain</td><td>NE Atlantic</td><td>13</td><td>200
(11 – 84)</td><td>Assess age-reading precision and improvement in age-reading precision of ages > 5 years</td><td>

Decrease in level of agreement for ALK readers
Strong bias found in age estimates of older fish
High level of agreement in locating first three annuli
Consensus ageing methods for ages > 5 years to be established at next workshop

<table>
<tr><th>Readers</th><th>Agreement (%)</th><th>APE (%)</th><th>CV (%)</th></tr>
<tr><td>All</td><td>47</td><td>41</td><td>45</td></tr>
<tr><td>ALK</td><td>57</td><td>24</td><td>32</td></tr>
<tr><td>Expert</td><td>57</td><td>21</td><td>29</td></tr>
</table>
</td></tr>
<tr><td>2004</td><td>Workshop</td><td>Belgium, Denmark, UK, France, Ireland, Portugal, and Spain</td><td>NE Atlantic</td><td>14
(70)</td><td>S1: 127
(11 – 59)
S2: 187
(10 – 58)
S3: 70
(14 – 87)</td><td>Try to establish ageing criteria for older fish; checking precision and relative bias of ALK readers; incorporation of new readers</td><td>

No agreed criteria for older fish
Difficult to maintain precision for fish older than 3 years
Confident age range dropped from 5 to 3 years from 2001 to 2003 exchanges, respectively
Studies on hake growth presented at workshop indicate that actual ageing criteria not accurate

<table>
<tr><th>Readers</th><th>Sample</th><th>Agreement (%)</th><th>APE (%)</th><th>CV (%)</th></tr>
<tr><td>All</td><td>S3</td><td>–</td><td>24 – 24</td><td>32 – 31</td></tr>
<tr><td>ALK</td><td>S1–S2</td><td>72 – 60</td><td>19 – 35</td><td>25 – 48</td></tr>
<tr><td>ALK</td><td>S3</td><td>–</td><td>20 – 15</td><td>25 – 20</td></tr>
<tr><td>Expert</td><td>S3</td><td>–</td><td>22 – 15</td><td>31 – 20</td></tr>
</table>
</td></tr>
</table>

2.2 Summary of the most recent (2004) hake otolith workshop report

Although the 2004 workshop report is available via the PGCCDBS document repository on the ICES website, we summarize it here because of the extent of the conclusions compared with previous workshop reports. See http://www.ices.dk/reports/acfm/pgccdbs/PGCCDBSdocrepository.asp

2.2.1 Introduction

In 2002, the WGHMM (ICES, 2003) recommended addressing hake ageing problems by means of (i) a hake otolith exchange programme (scheduled for 2003) focusing mainly on older fish and (ii) a subsequent workshop to discuss the results (scheduled for 2004). The results of the 2003 otolith exchange programme indicated a decrease in the precision of age readings compared with the precision of the 2001 exchange and a strong bias in the estimates for older fish.

The main goals of the 2004 workshop were to increase the precision of European hake otolith age estimation reached in previous exchanges (in 1997, 1999, 2001, and 2003) for ages up to 5 years and to improve the confidence in age estimation for older age groups (> 6 years).

The initial terms of reference (ToRs) for the 2004 workshop were changed as a result of recent advances reported in age and growth estimation from tagging experiments. The revised ToRs were:

- discussion of the results of the fourth otolith exchange (2003)
- discussion of new information regarding the estimation of age and growth and alternative methods of obtaining age–length keys (ALKs) for assessment purposes (e.g. elaboration of synthetic ALKs)
- checking the precision and bias of age readers involved in stock assessment
- discussion of age-reading criteria and problems identified for young and/or old fish
- incorporation of new readers in hake age estimation

2.2.2 Material and methods

The workshop was held from 18 to 20 October 2004 in Vigo (Table 2.1.1). A subset of 70 otoliths (S3) from an exchange of the 2003 collection was read in this workshop in the absence of information on fish length. This subset was selected to include otoliths that represented the full range of agreement obtained in the previous exchange (2003).

At the 2003 exchange, an ageing protocol was circulated together with a CD-ROM containing digitized images from otolith sections and the adopted ageing criteria (Piñeiro and Saínza, 2003). All readers were asked to measure the distances between the nucleus and (i) the first three translucent rings considered to be *annuli*, (ii) the check ring, and (iii) the edge. These measurements were marked on the images. This was to determine whether readers were counting the same translucent rings. In order to make analytical comparisons, the readers were split into three groups based on their level of experience: ALK readers, expert readers, and all readers.

Digital images, projected on a screen using a computer running TNPC Image Analysis Software, were used to facilitate individual interpretations and discussions between the readers.

Two analyses were undertaken:

1) comparison of readings from the 2001 exchange (S1) and the 2003 exchange (S2) for ALK readers, excluding otoliths from fish larger than 60 cm, in order to assess reader bias
2) comparison of the age-reading results from a subset of 70 otoliths (S3) with the 2003 exchange collection (S2; Table 2.1.1, last row)

The workshop analyses of the age-reading results were performed using the "Guidelines and Tools for Age Reading Comparisons" (Eltink *et al.*, 2000). Several indices and tests were used to assess the age-reading consistency of several readers from the same calcified structures, as recommended by Campana (2001):

1) **The percentage of agreement** is the ratio (as a percentage) between the number of coincident readings and the total number of readings and is one of the simplest methods available. However, Beamish and Fournier (1981) recognized that this percentage did not take into account the absolute age of the fish. Coefficient of variation (CV) and average percentage error (APE) are now preferred for the study of age-reading precision.
2) **The average percentage error** (APE; Beamish and Fournier, 1981) is an index of reading precision that is very useful for comparing series of observations. It is defined as

$$APE = \frac{100}{n}\sum_{i=1}^{n}\left(\frac{1}{r}\sum_{j=1}^{r}\frac{|x_{ij} - \bar{x}_i|}{\bar{x}_i}\right)$$

where n is the number of otoliths, r is the number of readings for each otolith, x_{ij} is the j value of age estimation for otolith i, and $\bar{x}_i$ is the mean age of otolith i. When averaged across many fish, it becomes an index of mean APE.

3) **The coefficient of variation** (CV) describes the precision errors in age reading by age group. It is statistically more robust and flexible than APE. It should be remembered that CV is very sensitive to low age values.

$$CV = \frac{100}{n}\left[\sum_{i=1}^{n}\left(\frac{\sigma}{\bar{x}_i}\right)\right]$$

where n is the number of otoliths, σ is the standard deviation for the otolith i, and $\bar{x}_i$ is the mean age of otolith i.

Because no validated ages are available, the reference age assumed is the modal age from readers who provide ALKs for stock assessment. In the case of bimodality, the estimate of modal age was based on data from the most expert readers.

The first three annual rings and check ring measurements were analysed using box-whisker plots by age and reader. Age-bias plots demonstrate both types of age-reading errors (precision and accuracy); however, because known-age material was not available, the bias in age reading was only assessed by precision.

Moreover, evidence from mark–recapture experiments, daily growth, and elaboration of synthetic ALKs (Annexes 6, 7, and 8 respectively of the 2004 workshop report; see Piñeiro *et al.*, 2004) were available and were presented during the workshop. This evidence gives insight into differential growth; thus, relevant results on growth contribute to the main conclusions that are crucial to assessment and management of the hake stocks.

2.2.3 Results and discussion

2.2.3.1 Comparison of results from the last two exchanges: 2001 (S1) and 2003 (S2)

The results revealed that, between the 2001 (S1) and 2003 (S2) exchanges, agreement dropped from 72 to 60%, whereas the APE increased from 19 to 35% and the CV increased considerably from 25 to 48%. The values obtained from the APE and CV indicated that precision of age estimation has decreased between both exchanges (Figure 2.1.1).

2.2.3.2 Comparison of results from the 2003 exchange (S2) and 2004 workshop (S3)

In general, the majority of readers tended to give lower ages for S3 than for S2. The ages assigned by readers to fish older than 3 years demonstrated a higher level of variability. The results indicated that it is difficult to maintain precision for fish older than 3 years (reference – or modal – age, not true age). In the S3 readings, the amplitude of confidence intervals generally decreased for all readers, and they tended to underestimate ages above 3 years (Figure 2.2.1).

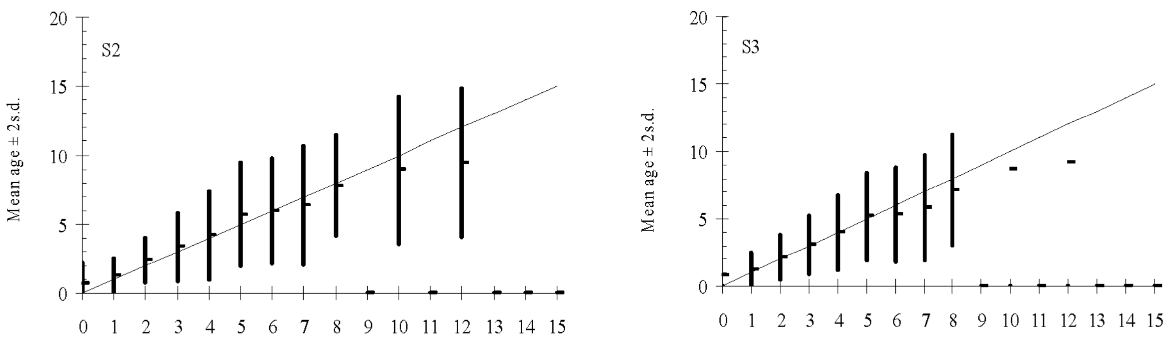

Figure 2.2.1. Age-bias plots for the same readers involved in the 2003 exchange (S2) and the 2004 workshop (S3). Mean age recorded ± 2 s.d. (standard deviation) are plotted against the modal age.

2.2.3.3 Analysis of fish less than 60 cm total length (S4)

The values obtained for APE and CV demonstrated hardly any improvement for the three groups of readers.

The radius measurement for the first three annual rings and the check ring indicated that all readers can clearly distinguish between these rings because similar median distances for these rings were obtained (Figure 2.2.2). This indicates the consistent nature of the ageing criteria for these first rings.

The classification of the otolith edge (opaque or translucent) was also discussed. Most of the confusion was caused by the frequent occurrence of a translucent edge through the whole year in young fish.

Considering the incorporation of new readers in hake age estimation, the results of this workshop demonstrate the difficulties in applying the established ageing criteria and reflect the importance of training in otolith age reading.

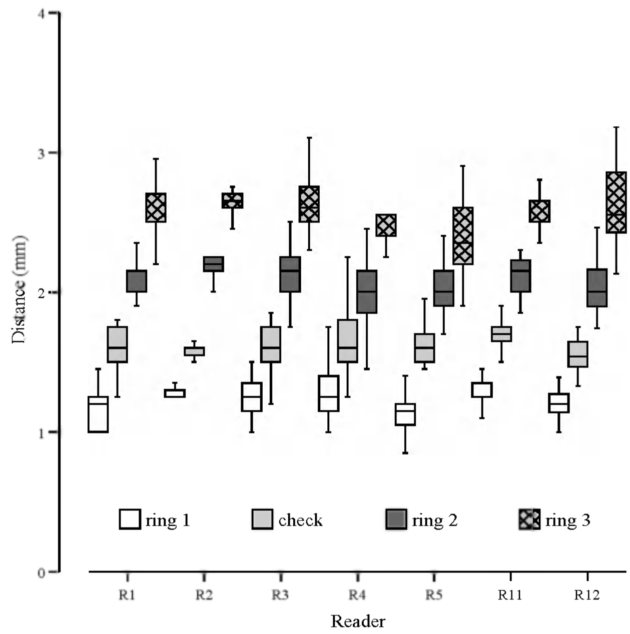

Figure 2.2.2. Ring measurement (mm) from the nucleus to the end of the translucent band for the following rings by all readers who provided stock-assessment age data: first annual ring (ring 1); check; second annual ring (ring 2); third annual ring (ring 3).

2.2.4 Conclusions

The ages assigned by readers to fish older than 3 years demonstrated a higher level of variability compared with those for younger fish. Taking into account the low precision obtained for those lengths, no agreed criteria were established for older fish.

The results of studies on hake growth presented at the 2004 workshop indicate that the ageing criteria are inaccurate.

The results from mark–recapture experiments provide evidence that ages are overestimated when using the internationally agreed criteria (de Pontual *et al.*, 2003).

2.2.5 Recommendations

Taking into account the loss of precision in the last exchange (2003) and workshop (2004), and the inaccuracy of ageing criteria evidenced by tagging experiments, workshop participants recommended that the supply of age-reading data for the elaboration of ALKs for stock assessment purposes should be interrupted in order to reallocate effort to the establishment of validated ageing criteria.

Validation studies are essential to the progress of hake ageing studies. Taking into account the recapture rate obtained in recent studies (de Pontual *et al.*, 2003), tagging is a very promising method for validating hake ageing.

In the meantime, the effort (for both time and people) that, until now, has been expended on the reading of otoliths should be allocated to other tasks, such as:

- tagging surveys, financed by national data collection and management programmes, in order to provide the reference material essential to the development of accurate age criteria that will yield reliable ALKs
- creation of a database of otolith images, weight, and other complementary biological information
- otolith microstructure studies (daily growth, etc.)

- length distribution analysis of surveys and available commercial catches
- research studies of the macrostructure pattern of otoliths in order to establish an understanding of the typology of the rings (annual rings and checks) and the mechanisms that control their deposition

Workshop participants recognized that these recommendations represent a turning point in the stock assessment of this species. In light of the results obtained in this workshop, as well as the recent advances in hake age validation (mark–recapture experiments, daily growth), it is important to avoid expending time in obtaining age data without assurance of its precision.

3 Summary of current knowledge

Over the years, many initiatives have been launched to address the problems of age determination of European hake. These have been reported in the scientific literature and very frequently in "grey literature", available in the public domain but not widely distributed. This section describes important recent studies that have contributed directly or indirectly to a better understanding of hake growth.

Initial attempts at studying the growth of European hake were made using mark–recapture methods (Belloc, 1935) and reading of scales (Birtwistle and Lewis, 1925; Belloc, 1929). Subsequently, most researchers have relied on the interpretation of rings in bony structures (otoliths) and on analyses of length–frequency distributions, such as modal progression analysis and the separation of modal components (Gulland and Rosenberg, 1992). Daily growth of this species has also been studied and has yielded important information about first-year growth (Morales-Nin and Aldebert, 1997; Arneri and Morales-Nin, 2000; Morales-Nin and Moranta, 2004; Kacher and Amara, 2005; Belcari *et al.*, 2006; Piñeiro *et al.*, 2008). Recent mark–recapture experiments are an important tool in improving the current status of knowledge of European hake growth. Although independent studies may indicate growth rates different from the ones inherent in standard age-reading criteria, mark–recapture results have been the driving force behind the general discussion on the hake fast-growth hypothesis.

Over the years, several techniques for making hake otoliths easier to examine have been employed (Christensen, 1964; Albrechtsen, 1968; Nichy, 1969) but even so, all of the researchers have encountered difficulties in interpreting the pattern of growth ring formation on otoliths of this species consistently. Therefore, studies on annual growth have reported very different growth estimates for hake in both the Northeast Atlantic and the Mediterranean Sea (Hickling, 1933; Bagenal, 1954; Meriel-Bussy, 1966; Robles *et al.*, 1975; Iglesias and Dery, 1981; Goñi, 1983; Goñi and Piñeiro, 1988; Guichet, 1988; Lucio *et al.*, 2000; Godinho *et al.*, 2001; Piñeiro and Saínza, 2003; ICES 2000, 2008a). Figure 3.1.1 shows the wide range in total length by age class and sexes combined. To minimize bias caused by different sample size and range, the estimated sample mean lengths-at-age are presented as the only comparable growth information available throughout the studies reviewed. It is important to assess whether such a variety of results is a consequence of diverse biological growth, an outcome of bias and inaccuracy in the age estimation methods, or a combination of both these factors.

3.1 Age-reading criteria

In order to standardize age-reading procedures, otolith preparation and ageing methods were agreed at two workshops conducted in 1997 and 1999 within the framework of the EU projects BIOSDEF (Anon., 1998) and DEMASSESS (Anon., 2000). The main achievement of these workshops was a set of internationally accepted interpretation criteria for hake otoliths up to the age of 5 years, allowing otolith readers from the various institutions involved in stock assessment to age hake consistently. The elaboration of these ageing criteria was a direct consequence of previous research studies on the growth pattern of hake otoliths, which were carried out by a group of experts on hake otolith patterns and biology. The agreed criteria rely on the experience obtained from previous hake ageing studies (Goñi and Piñeiro, 1988; Piñeiro and Hunt, 1989; Piñeiro *et al.*, 1992; Piñeiro and Pereiro, 1993). Additional information was also employed, namely length–frequency distributions from research and discard surveys (Piñeiro *et al.*, 1992; Pérez *et al.*, 1996). However, these criteria were never validated (Piñeiro and Saínza, 2003).

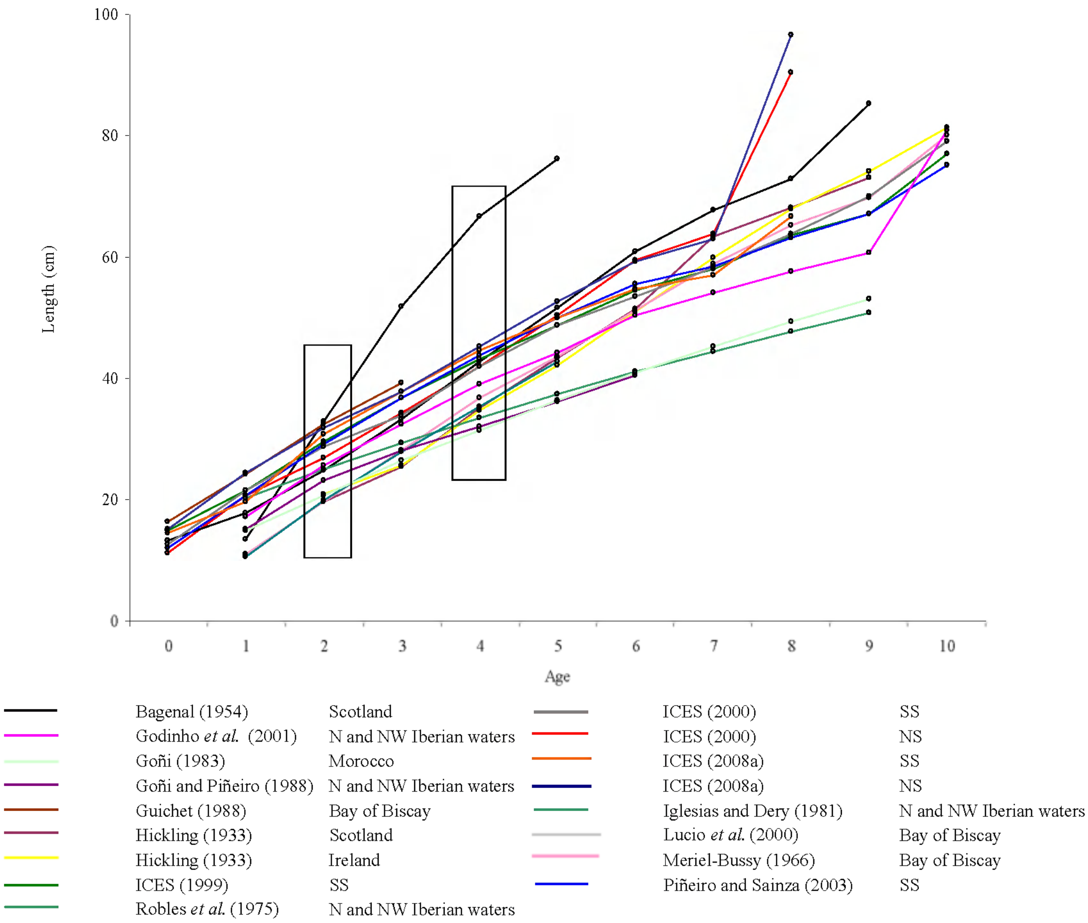

Figure 3.1.1. Mean length-at-age (sex combined) obtained by mean otolith age readings from different authors in Northeast Atlantic waters (NS, northern stock; SS, southern stock).

The standard age estimation criteria are derived from hake otolith sections, which have a concentric pattern of translucent and opaque bands around the nucleus when viewed under reflected light. One annual growth zone, or *annulus*, consists of one opaque and one translucent ring or band. Counts of annual translucent rings, preferably on the ventral region, are used to estimate ages. The first annual ring (1) is identified despite the presence of checks (−3, "larval"; −2, "pelagic"; −1, "demersal") around the nucleus (Figure 3.1.2). Growth during the first and second years is great compared with subsequent annual increments. Although the position of the first annual ring varies, its recognition is aided by the frequent presence of a well-marked translucent ring along the dorso-ventral axis of the section between the two first annual bands. This check is frequently found on the section otolith at ca. 1.57 mm (mean value ±0.13, $n = 608$) from the nucleus. The pattern of otolith growth presents two translucent rings per year during the first 3 years of life and thereafter only one, which has been associated with the process of sexual maturation.

Classification of the otolith edge type (translucent or opaque) tends to be complicated by the high incidence of false rings. Translucent edges appear year-round. On average, more than 60% of the otoliths presented translucent edges, indicating a high incidence of checks, particularly in summer (Piñeiro and Saínza, 2003). In general, two peaks of translucent edges per year were observed. The most important peak was in winter (November) and the secondary peak was in spring–summer (April–June). Nevertheless, it is assumed that otolith edges follow the predominant pattern of translucent-in-winter and opaque-in-summer; by convention, an otolith with a translucent edge is not considered to be one year older until 1 January of every year.

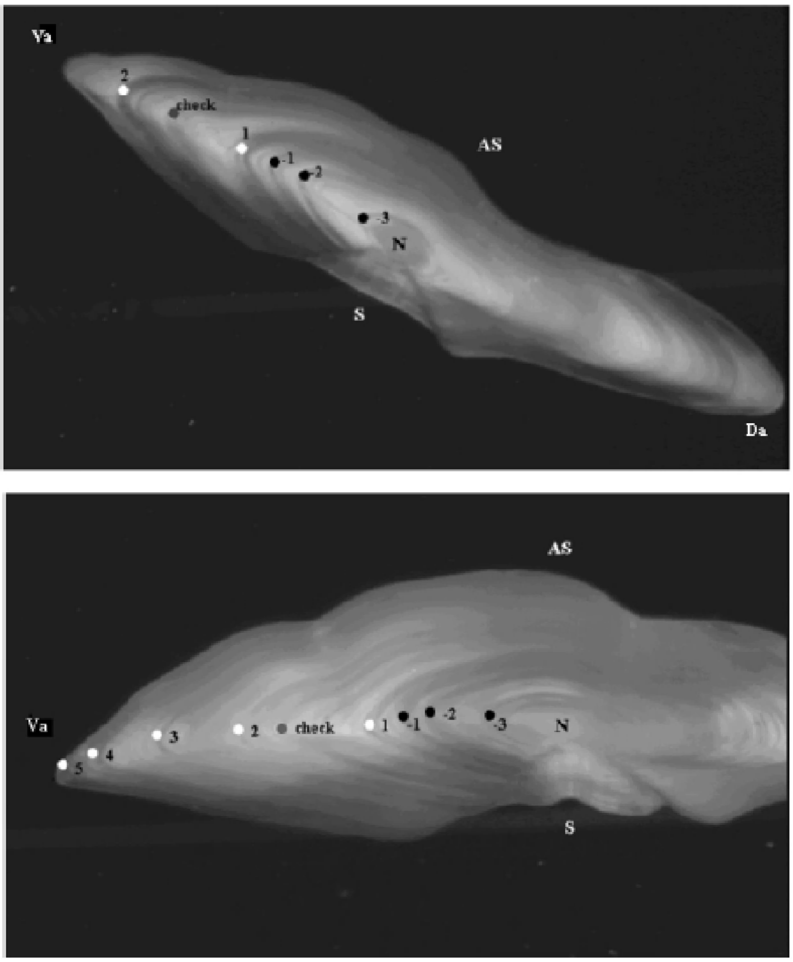

Figure 3.1.2. Transverse sections of *sagitta* otoliths of two hake specimens collected in Iberian Atlantic waters in 1997. Top: a male of 30 cm total length caught in October and aged 2 years. Bottom: a female of 47 cm total length caught in April and aged 5 years. Photographs (× 20, reflected light) show central area around the nucleus (N) where three false rings (–1, –2, and –3) are visible before the first *annulus* and one (check) between the first and the second *annulus* (1 and 2). Annual rings are visible as translucent zones (white dots). (Da = dorsal apex; Va = ventral apex; S = sulcus; AS = antisulcus; from Piñeiro and Saínza, 2003).

3.2 Reproductive behaviour

One of the key issues in relating length and age for hake is the extended spawning period. Spawning of hake in the Northeast Atlantic has been studied by several authors (Lucio *et al.*, 2000; Piñeiro and Saínza, 2003; Lannin, 2006; Murúa, 2006; Murúa *et al.*, 2006; Domínguez-Petit, 2007). Results from these studies indicate that spawning takes place over an extended period and occurs earlier in more southerly latitudes and progressively later towards the northern latitudes. It is possible to find mature specimens throughout the year, but prolonged spawning is more evident in females (Murúa and Motos, 2006; Table 3.2.1). Spawning is one of the more important biological aspects that will be reflected in the otolith growth patterns if this species registers the energetic effort of spawning as rings in the otoliths (Morales-Nin and

Moranta, 2004). In the Mediterranean Sea, Oliver (1991) also reported a long spawning season for this species, with peaks in spring and autumn demonstrating a marked interannual variability related to individual growth variability. Morales-Nin *et al.* (1998) also reported that the ring pattern depends on sex and sexual activity.

The European hake is a batch-spawner with indeterminate fecundity. Reproductive modality of this species is highly asynchronous, both at the individual level, namely oocyte development, and at the population level, namely spawning pattern (Murúa, 2006; Domínguez-Petit, 2007). These fluctuations may increase the chances of survival of the offspring and decrease the natural mortality of the cohorts (Murúa *et al.*, 1996). The extended spawning season also indicates the occurrence of individuals of very different sizes belonging to the same year class (born in the same calendar year). These size differences will be greater with faster growth rates. This factor will always be an obstacle to accurate age estimation for hake.

A study of whole and sectioned hake otoliths from Galician waters and the Cantabrian Sea, based on ring measurement analysis, found that otolith ring pattern was a function of hatch date (Piñeiro and Hunt, 1989). So, for average total length at age 1 (which can vary from 10 to 20 cm), two extreme otolith types were evident, corresponding to early and late hatching.

In relation to the spawning areas, available studies demonstrate that hake migrate to spawn, although spawning behaviour appears to vary with latitude. Belloc (1935) and Hickling (1933) observed that mature adults concentrate in deep waters but spawn in shallower waters. Males reach first maturity at a shorter length and younger age than females. Therefore, in the Northeast Atlantic, the northern stock has a mean length and age at first maturity of ca. 39 cm and 3.4 years for males and 47 cm and 4.2 years for females (Lucio *et al.*, 2000; ICES, 2007a). For the southern stock, Piñeiro and Saínza (2003) reported ca. 33 cm and 2.5 years for males and 45 cm and 4.4 years for females. Males grow slightly faster than females up to age 2 years, but from age 3 years onward, female growth rates surpass those of males. Females also grow to larger sizes and live longer than males. The difference in growth rate coincided with the onset of sexual maturity.

Table 3.2.1. Hake spawning season and their main peaks found by study (dark grey = main peak of the spawning season; light grey = peak of the spawning season).

Area	Study	Month											
		1	2	3	4	5	6	7	8	9	10	11	12
Bay of Biscay	Lucio *et al.* (2000)	░	■	■	░	░							
Bay of Biscay	Álvarez *et al.* (2004)	░	■	■	░	░							
Bay of Biscay	Murúa and Motos (2006)	■	■	■	░	░	░	░	░	░	░	░	░
Celtic Sea	Fives *et al.* (2001)				■	■	■						
Celtic Sea	Lannin (2006)	░	░	░	░	░	░	░	░	░			
Galician waters and Cantabrian Sea	Alcázar *et al.* (1983); Pérez and Pereiro (1985)	░	■	■	░								░
Galician waters and Cantabrian Sea	Piñeiro and Saínza (2003)	░	■	░	░	░							░
Galician waters	Domínguez-Petit (2007)	■	■	■	░	░	■	■					
Gulf of Cadiz	Silva *et al.* (2007)	░	░			░	░	░	░	░			
Mediterranean Sea	Oliver (1991)			■	■	■				■	■	■	░
Adriatic Sea	Arneri and Morales-Nin (2000)						■	■	■	░	░	░	░
Mediterranean Sea	Morales-Nin and Moranta (2004)						░	░	░	■	■	■	░
Greek waters	Mytilineou and Vassilopoulou (1988)	░	░	░	░	░	░	░	░	░	░	░	░

3.3 Length–frequency analysis

The analysis of length–frequency distributions can provide useful estimates of relative age and growth of fish. Several studies have been carried out using length–frequency distributions of hake in the Atlantic (Hickling, 1933; Bagenal, 1954; Guichet, 1988; Piñeiro and Pereiro, 1993; Godinho *et al.*, 2001) and the Mediterranean (Orsi-Relini *et al.*, 1989; Recasens, 1992; Alemany and Oliver, 1995; Morales-Nin and Aldebert, 1997; García-Rodriguez and Esteban, 2002). The most commonly used length-based method is the relationship between otolith radius and fish size (Goñi, 1983; Goñi and Piñeiro, 1988; Piñeiro and Hunt, 1989; Recasens, 1992; Table 3.3.1). However, the extended spawning period and the influx of new recruits throughout the year make the length-progression analysis difficult to interpret because modal age groups may not be distinguished and/or more than one mode may belong to the same year class (Goñi, 1983; Piñeiro and Hunt, 1989; Godinho *et al.*, 2001; Domínguez-Petit, 2007).

Table 3.3.1. Overview of European hake length–frequency analysis studies: growth rate in first year and total length (TL) for age 1 by author, area, and method.

Study	Area	Growth rate, first year, (cm month^{-1})	TL at age 1 (cm)	Method
Hickling (1933)	West of Ireland		10.5	Petersen (1891)
Bagenal (1954)	Scotland		22.3	Petersen (1891)
Goñi (1983)	NW African Shelf		13 - 16.4	Backcalculation
Goñi and Piñeiro (1988)	Galician waters and Cantabrian Sea		15	Backcalculation
Guichet (1988)	Gulf of Biscay		24.1	NORMSEP
Piñeiro and Hunt (1989)	Galician waters and Cantabrian Sea		10 - 20	Backcalculation
Orsi-Relini *et al.* (1989)	Ligurian Sea	0.8 - 1.2 (spring) 0.7 - 0.9 (autumn)		Modal progression analysis
Recasens (1992)	Catalan Sea	1.1		Modal progression analysis
Piñeiro and Pereiro (1993)	Galician waters and Cantabrian Sea		20	Petersen (1891)
Alemany and Oliver (1995)	Balearic Sea	1.8 (only females)		Bhattacharya (1967) FISHPARM software package
Morales-Nin and Aldebert (1997)	Gulf of Lion	1.15	14.7	MIX (MacDonald and Pitcher, 1979)
Godinho *et al.* (2001)	Portuguese continental waters		14.4 - 21.8	Bhattacharya (1967) FiSAT software package

3.4 Growth parameters

The literature on European hake growth abounds with estimates of mean lengths-at-age and growth parameters obtained for different geographic regions using a variety of ageing methodologies. Almost all studies on hake growth deal with fitting von Bertalanffy (1938) or other growth functions to length-at-age data, where the age data are inferred either from calcified structures or by length–frequency distributions. They are difficult to assess and compare because their estimation depends greatly on the size and age ranges covered in the study. Very few of these studies use validated ages. A summary of growth parameters estimated by sex, both separately and combined, is presented in Table 3.4.1. It is difficult to make consistent comparisons because of differences in the sampling design, age estimation method used (e.g. otolith age estimation, length–frequency analysis), calcified structure (e.g. otoliths, scales) and parameter estimation procedures (e.g. assumed asymptotic length). The wide range of results may be a consequence of the methods used for age estimation (bias/inaccuracy) or attributable to biological factors, such as the complexity of otolith pattern.

Table 3.4.1. Growth parameters obtained by von Bertalanffy growth function (K, L_∞, and t_0) in different studies by sex (m = males; f = females; c = combined sexes), age estimation method, stock (M = Mediterranean Sea; N = Atlantic northern stock; S = Atlantic southern stock), and geographic area.

Study	Sex	Method	Stock	Area	K (cm year^{-1})	L_∞ (cm)	T_0 (year)
Belloc (1929)	c	Scales	–	Africa	0.078	56.6	−0.586
Hickling (1933)	c	Otoliths	N	S Ireland	0.087	128.6	–
Bagenal (1954)	c	Petersen	N	Scotland	0.710	126.4	–
	c	Otoliths			0.204	125.9	–
Meriel-Bussy (1966)	c	Otoliths	N	Bay of Biscay	0.059	171.8	–
Guichet *et al.* (1973)	m	Otoliths	N	NE Ireland	0.069	124.0	–
	f				0.070	124.0	–
Guichet *et al.* (1974)	m	Otoliths	N	NE Ireland	0.024	268.2	–
	f				0.087	123.7	–
Robles *et al.* (1975)	c	Otoliths	S	Galician waters and Cantabrian Sea	0.070	125.4	–
Descamps and Labastie (1978)	m	Otoliths	N	Bay of Biscay	0.148	83.0	−0.420
	f				0.098	116.0	−0.510
Iglesias and Dery (1981)	c	Otoliths	S	Galician waters and Cantabrian Sea	0.060	99.9	−2.740
	m				0.150	63.4	−1.030
	f				0.060	99.8	−2.360
Goñi (1983)	c	Otoliths/ Back-calculated	S	NW African Shelf	0.064	110.0	−0.760
	m				0.067	100.0	−1.090
	f				0.065	110.0	−0.820
ICES (1991)	c	Otoliths	S	Galician waters and Cantabrian Sea	0.080	100.0	−1.420
Alemany *et al.* (1993)	f	FISHPARM/ Bhattacharya	M	Balearic Sea	0.214	113.2	0.311
ICES (1993)	c	Otoliths	N	Bay of Biscay	0.073	127.5	−1.130
Aldebert and Recasens (1995)	m	FISHPARM/ Bhattacharya	M	Gulf of Lion	0.149	72.8	−0.383
	f				0.124	100.7	−0.350
Alemany and Oliver (1995)	c	FISHPARM/ Bhattacharya	M	Balearic Sea	0.184	126.9	0.035
García-Rodriguez and Esteban (1995)	c	Petersen	M	Mediterranean Sea	0.123	113.2	0.137
Lucio *et al.* (2000)	c[1]	Otoliths	N	Bay of Biscay	0.124	110.0	−0.452
	m[1]				0.181	80.0	−0.724
	f[1]				0.122	110.0	−0.619
Godinho *et al.* (2001)	c	Otoliths	S	Portuguese continental waters	0.08	110	−0.97
García-Rodriguez and Esteban (2002)	m	FiSAT (subp. ELEFAN)	M	Mediterranean Sea	0.20	93.0	−0.091
	f				0.21	108.0	0.115
	c				0.21	108	0.115
Piñeiro and Saínza (2003)	c[1]	Otoliths	S	Galician waters and Cantabrian Sea	0.07	120.5	−1.72
	c				0.13	88.01	−1.17
	m				0.18	70.06	−0.97
	f				0.13	89.04	−1.15
de Pontual *et al.* (2006)	c	Mark – recapture	N	Bay of Biscay	0.362	89.9	–
	c[1]				0.250	110	–
	m[1]				0.436	80	–
	f[1]				0.261	110	–

[1] L_∞ fixed.

3.5 Bioenergetics studies

In the Northeast Atlantic, the hake is an ichthyophagous species. Adults feed on fish, mainly blue whiting, other gadoids, and pelagic fish, whereas juveniles prey mainly upon planktonic crustaceans (mainly euphausids, copepods, and amphipods). Cannibalism of juveniles by adults has also been reported (Velasco and Olaso, 1998).

In Galician waters and the Cantabrian Sea, the hake is one of the top predators of the demersal community and occupies one of the highest trophic levels (Velasco *et al.*, 2003). The variability in diet depends mainly on two factors: length and depth (Velasco, 2007). Velasco (2007) indicated an important change, which occurs at a fish length of ca. 18 cm, from a benthic diet based on crustaceans to a pelagic and mesopelagic diet based on small fish. This change may be responsible for the characteristic check found in the ring pattern of hake otoliths described by several authors (Goñi and Piñeiro, 1988; Piñeiro and Hunt, 1989). The presence of this check (ca. 1.5 mm; see Section 3.1), which appears to be well marked in the otolith (Piñeiro and Saínza, 2003), may be linked to some biological or behavioural event, such as a dramatic change in diet. Velasco and Olaso (1998) noted that, in the Calabrian Sea, the diet of hake between 15 and 20 cm in length changed from 88% invertebrates to 97% fish. This might explain the presence of the check because it appears to be independent of season and occurs when the fish attain ca. 19 cm in length (Goñi and Piñeiro, 1988).

A study conducted by Riis-Vestergaard *et al.* (2000) on the rate of food consumption by hake, with estimates based on a bioenergetic model and gastric evacuation methods, revealed that the growth rate may be underestimated. Although bioenergetics studies do not permit accurate age estimation, they can be used as an external support for age estimation.

3.6 Daily growth

The study of daily growth increments is a useful tool for the examination of growth and length-at-age of young fish (Pannela, 1971; Morales-Nin and Aldebert, 1997). This methodology has been indirectly validated in larvae and juvenile European hake in the Adriatic Sea (Arneri and Morales-Nin, 2000) and in experimentally reared conditions in Norway (Morales-Nin *et al.*, 2005). Otolith microstructure analysis has been used to estimate juvenile growth rates and to determine the length corresponding to the first year of life of hake in different areas of the Mediterranean Sea (Morales-Nin and Aldebert, 1997; Arneri and Morales-Nin, 2000; Morales-Nin and Moranta, 2004; Belcari *et al.*, 2006) and in Atlantic waters (Kacher and Amara, 2005; Piñeiro *et al.*, 2008). This method was also used to study larval growth and development, and mortality rates, in both the Atlantic (Álvarez and Cotano, 2005; Bjelland and Skiftesvik, 2006) and the Mediterranean (Palomera *et al.*, 2005).

Hake growth during the first year of life is characterized by the occurrence of different check rings in the otolith. These are probably related to physiological and environmental events and make it difficult to locate the first annual translucent ring (Morales-Nin *et al.*, 1998; Piñeiro and Saínza, 2003). Erroneous interpretation of these rings could be the source of the differences in the length estimated in the first year in several studies (Morales-Nin *et al.*, 1998; Piñeiro and Saínza, 2003). Morales-Nin and Aldebert (1997) noted that, in the Gulf of Lion, hatching occurred throughout the year, indicating that irregular growth during the first year of life may be related to seasonal variations in the times of hatching and recruitment to the bottom producing different growth rates. Morales-Nin and Moranta (2004) found hake recruits year-round in the Mediterranean Sea, with modes between 7 and 11 cm total length (TL), depending on the season. Otolith analysis indicated monthly variations in growth rates (1.2–2.5 cm m^{-1}), with a maximum in summer corresponding to fish hatched the previous autumn. Arneri and Morales-Nin (2000) studied the early development of hake otoliths and obtained a mean total length of 15 cm at the end of the first year, with monthly variable growth rates (1.1–1.6 cm month^{-1}). Kacher and Amara (2005) note that, in age 0 hake (age < 365 d) in the Bay of Biscay and Celtic Sea (northern stock area), two hyaline rings were observed and, in one individual of 22.5 cm, a third ring corresponding to 329 d from the accessory growth centre was detected. They found that juvenile hake reach daily growth rates ranging from 0.71 mm d^{-1} to 0.74 mm d^{-1}. These results suggest that juvenile hake growth rates are faster than previously thought for this species (Table 3.6.1) and are consistent with the growth rates from tagging studies. The size at first year estimated by Kacher and Amara (2005) is close to the size when hake lay down their second-year ring (age group 1), according to the age data obtained by Descamps and Labastie (1978), Goñi and Piñeiro (1988), and Piñeiro and Saínza (2003).

A daily growth study using hake otoliths from the northwest Mediterranean obtained a mean total length of 18.3 cm at the end of the first year of life and a mean growth rate of ca. 1.5 cm month^{-1} (Belcari *et al.*, 2006). A new approach to the estimation of daily growth in Atlantic juvenile hake, based on both sagittal and transversal otolith sections (Piñeiro *et al.*, 2008), found that, by the end of the first year, hake from northwest Iberian waters may attain a length of 25.3 cm, demonstrating an average individual growth rate of 0.66 mm d^{-1} ± 0.06 (Table 3.6.1). These authors observed that the transversal section used to study the juvenile phase presented a particularly clear sequence of microincrements which, at macroscopic scale, consisted of successive wide opaque zones (OZ) and thinner translucent zones (TZ), as depicted in Figure 3.6.1. The latter zones correspond either to seasonal growth structures or to fish-specific responses to endogenous or environmental factors (Courbin *et al.*, 2007).

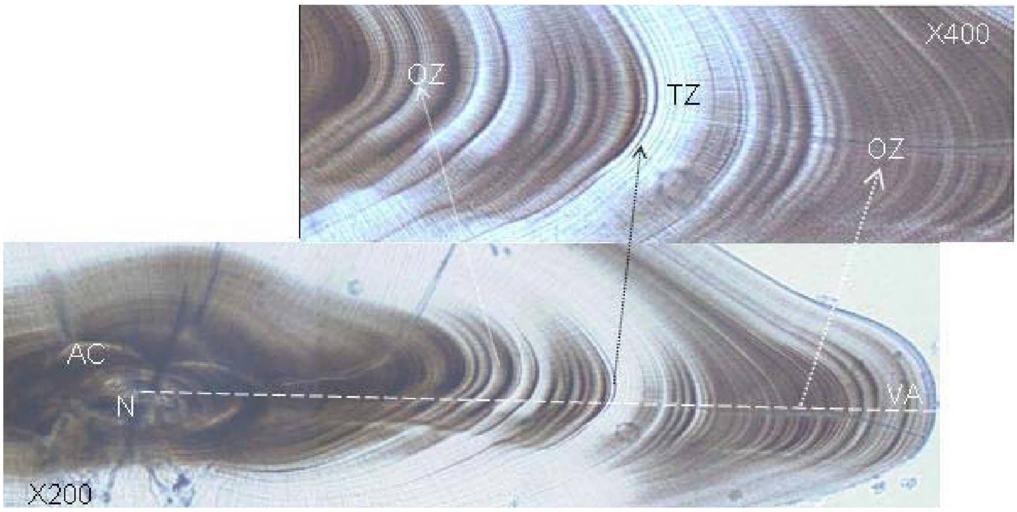

Figure 3.6.1. Bottom: hake otolith, sectioned on the transversal plane (under transmitted light, × 200 magnification), demonstrating a central zone with the nucleus (N) and accessory-growth centres (AC). The daily growth increments visible on the ventral axis (VA) are used for counting. Top: detail of a sequence (× 400 magnification) of growth increments grouping in the translucent (TZ) and opaque zones (OZ) and demonstrating the variation of increment widths that indicates seasonal growth variations.

Growth studies based on otolith daily age estimates have produced great discrepancies in the estimated growth rates throughout the juvenile phase and, consequently, in the size attained at the end of the first year of life. The highest daily growth rates (2.16–2.22 cm month^{-1}) were found in Atlantic hake (Kacher and Amara, 2005), which would result in a juvenile total length of 23.8 cm at the end of the first year. Lowest growth rates were observed in Mediterranean hake (Table 3.6.1). Nonetheless, it must be stated that these differences may be the consequence of different methodological approaches, geographical location, and seasonal origin of the samples.

Table 3.6.1. Overview of hake daily growth data obtained by various studies: growth rate (GR), mean length (ML) at first year of life, number of otoliths sampled, total length (TL) range, and study area.

STUDY	GR (CM MONTH $^{-1}$)	ML AT FIRST YEAR OF LIFE (CM)	NO. OTOLITHS	TL (CM)	AREA
Morales-Nin and Aldebert (1997)	1.15	16	81	10.5–20.7	Gulf of Lion
Arneri and Morales-Nin (2000)	1.1–1.6	15	145	1.6–16.4	Central Adriatic
Morales-Nin and Moranta (2004)	1.2–2.5	–	153	2.5–25	Mediterranean Sea
Kacher and Amara (2005)	2.16–2.22 * (0.72–0.74 mm d^{-1})	23.8	107	6–22	Bay of Biscay and Celtic Sea
Palomera *et al.* (2005)	0.45–0.57 * (0.15–0.19 mm d^{-1})	–	71	0.25–0.91	Mediterranean Sea
Álvarez and Cotano (2005)	0.45–0.51 * (0.15–0.17 mm d^{-1})	–	–	0.2–1.2	Bay of Biscay
Morales-Nin *et al.* (2005)	1.8	–	1	13.5	Rearing conditions
Belcari *et al.* (2006)	1.3–1.7	18.3	579	4–20	Tyrrhenian Sea
Bjelland and Skiftesvik (2006)	1.8	–	–	–	Rearing conditions
Piñeiro *et al.* (2008)	1.98 ± 0.18 * (0.66 ± 0.06 mm d^{-1})	25.2	103	3–25	Galician waters and Cantabrian Sea

* Data calculated from original study.

3.7 Mark and recapture

Age determination of hake is accompanied by two main sources of error: accuracy and/or precision (Campana, 2001). Marking (tagging) and recapturing fish is one of the best methods available for validating the periodicity of growth increment formation. Validation studies on age estimation for hake based on tagging were only accomplished very recently because of the fragility of the species and its bathymetric distribution (Lucio *et al.*, 2000; de Pontual *et al.*, 2003; Piñeiro *et al.*, 2007). An early experiment carried out by Belloc (1935) recovered only one individual, with a growth rate of 16.7 cm year^{-1} after eight months at sea. Lucio *et al.* (2000) reported three recaptures from 151 released tagged fish, but only one fish was retrieved after a very short time at liberty (23 d).

Results from tagging experiments (de Pontual *et al.*, 2006) conducted in the Bay of Biscay demonstrated that the somatic growth of the recoveries was double that expected from published von Bertalanffy growth functions (VBGFs) for the species (Figure 3.7.1). The growth rate, when restricted to recoveries that had spent at least one summer and one winter at liberty, was estimated to be 19.82 ± 1.49 cm year^{-1} ($n = 6$). Growth underestimation was related to age overestimation, which was demonstrated by two independent analyses: (i) results based on a blind interpretation of marked otoliths by two experts involved in routine hake age estimation demonstrated that the age estimates were neither accurate nor precise (Figure 3.7.2); (ii) the predicted otolith growth was inconsistent with the observed otolith growth (Figure 3.7.3). Both types of otolith analyses invalidated the international agreed age-estimation method and demonstrate the need for further research (de Pontual *et al.*, 2006).

Results from a tagging experiment conducted off the northwest Iberian Peninsula indicated that hake growth rate, within the total length range studied, is about double that derived by the standard agreed criteria of otolith age estimation (Piñeiro and Saínza, 2003). Results from this work support the fast-growth hypothesis of this species (Figure 3.7.4). The results indicate that European hake would reach a total length of ca. 25, 45, and 60 cm at the end of the first, second, and third years of life, respectively, instead of 20, 29, and 37 cm, as estimated using the standard agreed criteria described by Piñeiro and Saínza (2003).

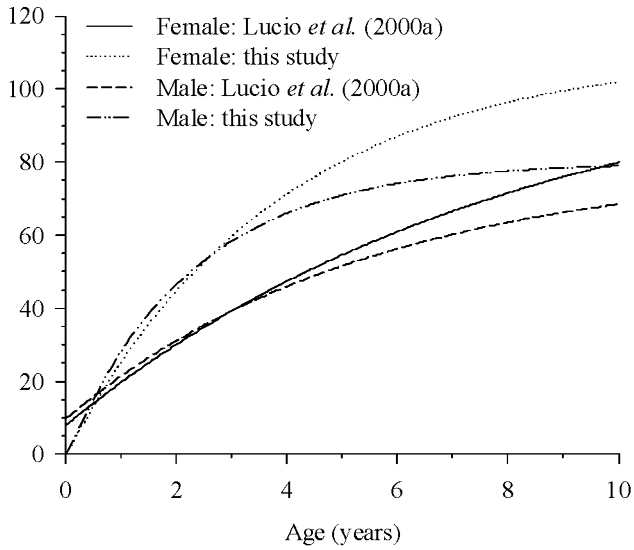

Figure 3.7.1. The von Bertalanffy growth function (VBGF) fitted from recapture data for male (dashed line) and female (solid line) hake and the corresponding VBGF given by Lucio *et al.* (2000); male (dotted and dashed lines), female (dotted line). Fitting was performed using fixed L_∞ values given by Lucio *et al.* (2000): 80 and 110 cm for males and females, respectively (from de Pontual *et al.*, 2006).

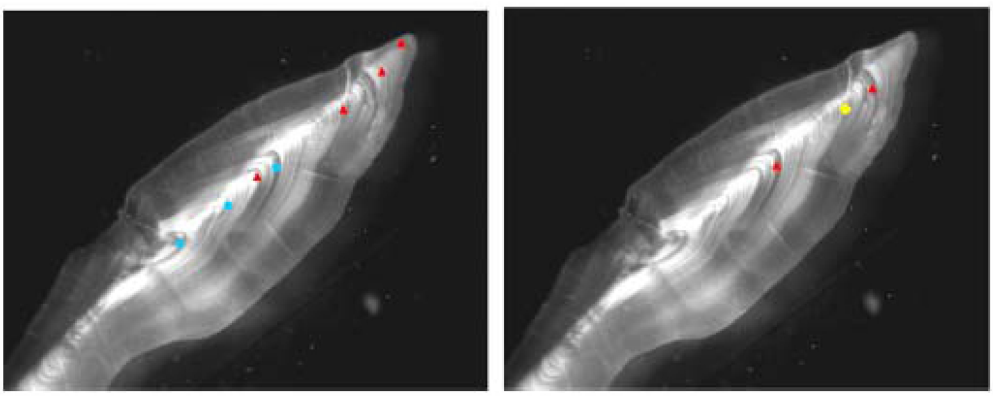

Figure 3.7.2. Transverse section of an oxytetracycline (OTC) marked otolith observed under reflected light. Left: blind interpretation (age 4+ years); right: new interpretation (age 2+ years). Blue indicates the false rings (FR) described by Piñeiro and Saínza (2003); red indicates the winter rings (WR); and yellow indicates the OTC mark. The fish was recaptured 301 d after tagging (TL at capture, 30 cm; TL at recapture, 49 cm; from de Pontual *et al.*, 2006).

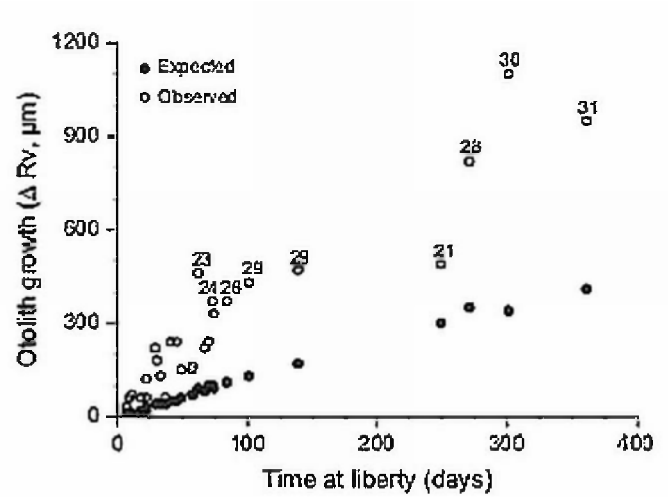

Figure 3.7.3. Comparative analysis of expected and observed otolith growth with respect to time at liberty. Otolith growth (ΔRv) is the distance (in mm) from the oxytetracycline mark to the edge, measured on the ventral radius. Numbers above the points indicate fish lengths (in cm) at tagging (from de Pontual *et al.*, 2006).

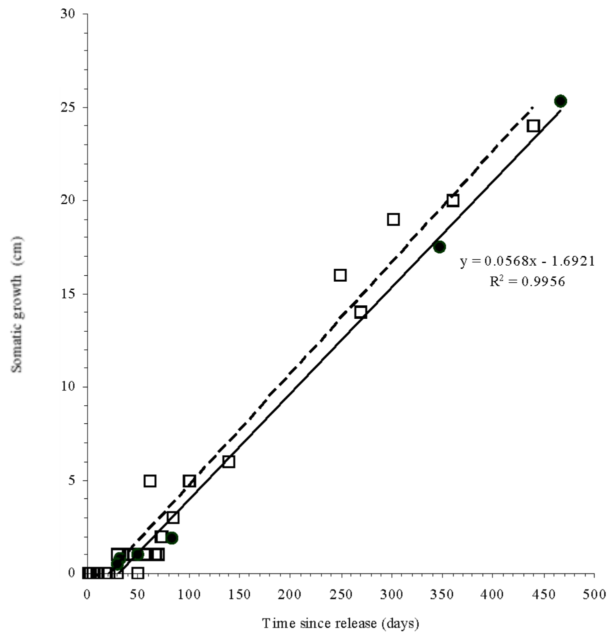

Figure 3.7.4. Somatic growth increment (total length, cm) as a function of time since release. Black dots and solid line, data from southern hake stock tagging experiment (Piñeiro *et al.*, 2007); squares and dashed line, data from northern hake stock tagging experiment (de Pontual *et al.*, 2006) restricted to comparable period (up to 500 d since release; from Piñeiro *et al.*, 2007).

Although earlier tagging studies had very low tag return rates (Belloc, 1935; Lucio *et al.*, 2000), their findings also indicated a much higher growth rate for this species (Table 3.7.1). The striking consistency between the results and somatic growth rates from tag recoveries of both northern and southern hake stocks (Piñeiro *et al.*, 2007) is in accordance with genetic studies, which do not find significant differences between the two stocks (Cimmaruta *et al.*, 2005).

Unfortunately, there are currently insufficient validated data to develop an alternative robust age estimation method for European hake. Direct measures of growth derived from mark–recapture experiments provide invaluable data for the resolution of the ageing problem.

Table 3.7.1. Summary of results of available mark–recapture studies of European hake (NS = northern stock; SS = southern stock; TL = total length; GR = growth rate).

Tagging experiment			Recapture results					
Study	Location Stock	No. released	No. tagged	TL release (cm) (mode)	Days since release	TL recapture (cm)	GR (cm d^{-1} mean ± s.d.)	Recapture rate %
Belloc (1935)	SW Ireland NS	78	1	28.9	255	40.6	0.046	1.3
Lucio *et al.* (2000)	South Bay of Biscay NS	152	1	56	24	60	0.166	1.9
de Pontual *et al.* (2006)	Bay of Biscay NS	1307	36	21–40 (29)	1-1066	24–67	0.038 ± 0.004	3.1
de Pontual *et al.* (2006)	Bay of Biscay NS	–	10*	21–33	101– 1066	34–67	0.052 ± 0.009	–
Piñeiro *et al.* (2007)	NW Iberian Peninsula SS	527	6	29–36 (29)	29–466	31–56	0.032 ± 0.016	1.3
Piñeiro *et al.* (2007)	NW Iberian Peninsula SS	527	2*	29-30	347–466	46.5–56	0.052 ± 0.003	–

* Fish that were >100 days at sea.

4 Key issues

The key issues concerning hake are representative of the main problems encountered in the assessment of many European stocks. A large number of otolith exchanges and workshops have been held, aimed at improving the consistency and precision of age reading for hake. Usually, the reports of these studies have not been published in refereed journals and have been available only in grey literature. However, much of the work provides useful insights into the problems of age determination in hake and, as a result, some of the key findings and issues raised from these reports are listed below. The report from the most recent workshop (in 2004, summarized in Section 2.2) provides detailed information on the problems encountered.

4.1 Complexity of the otolith growth pattern

The interpretation of otolith growth marks is often a difficult task in which subjectivity increases with the complexity of the structural pattern of the otolith, as is the case for hake (Courbin *et al.*, 2007). Improved understanding of the biological meaning of growth marks and the development of a quantitative framework aimed at defining an objective procedure for otolith interpretation are complementary solutions to reducing interpretation subjectivity, and are especially needed for complex species (Courbin *et al.*, 2007).

Macrostructures visible on otoliths may have different origins. The identification of these macrostructures and the establishment of the link between the macrostructure and an event in the life cycle represent a fundamental step in accurate age estimation. Hake otolith interpretation is complex because of the presence of many macrostructures. There are many thin translucent zones (TTZs) that probably correspond to short environmental and/or physiological events, and the difficulty in interpreting such otoliths often increases with the size of the fish (Courbin *et al.*, 2007).

Goñi and Piñeiro (1988) found an average number of 11 translucent rings laid down in hake otolith sections of individuals within the length range 32–38 cm. If it is assumed that fish of that size were 3–5 years old (according to the standard age-reading criteria described by Piñeiro and Saínza, 2003), this would mean six to eight false rings in addition to the true annual rings.

The complexity of hake otolith macrostructure and growth variability is believed to be related to the long spawning season (Casey and Pereiro, 1995; Anon., 1998; Domínguez-Petit, 2007). The almost continuous recruitment throughout the year, as a result of multiple spawning, causes variable ring patterns. The sexual dimorphism in growth rates and different ring patterns, depending on the hatching time and geographical area, render otolith interpretation more difficult and contribute to the discrepancies that exist between different otolith readers. Piñeiro (2000) noted that the major difficulties in otolith age estimation were (i) the location of the first annual ring (first *annulus*), (ii) the classification of the rings as annual or false, and (iii) the interpretation of the edge. The identification of the first annual ring is problematic because of the presence of several checks associated with juvenile growth (Table 4.1.1).

Piñeiro and Saínza (2003) observed that these difficulties could be related to some generally well-marked false rings that very frequently occur before, during, or after the first *annulus* deposition. This makes it very difficult to identify the first annual ring and constitutes the most important source of uncertainty in hake otolith age reading. The wide range of length-at-age estimates given by different authors for European hake in 2003 was also an issue.

Casey and Pereiro (1995) held similar views and concluded that there is a large number of different descriptions of European hake growth, depending on the area of capture and on the interpretation of the rings on the otoliths. However, it was impossible at that time to determine whether the reported differences were real or the result of difficulties in age determination.

Fariña and Abaunza (1991) considered that the growth interruptions marked on hake otoliths could be related to depth stratification in relation to size and age. The predatory and reproductive behaviours of hake result in seasonal movements through habitats with different environmental conditions (Fariña and Abaunza, 1991).

Table 4.1.1. False ring distances (backcalculated) identified in hake otoliths before the first annual ring in different studies.

STUDY	GEOGRAPHICAL AREA	FALSE RING DISTANCES (CM)
Descamps and Labastie (1978)	Bay of Biscay	7 and 12
Iglesias and Dery (1981)	Galician waters and Cantabrian Sea	Three rings (<12)
Goñi (1983)	NW African Shelf	2, 4, 7, and 10
Goñi and Piñeiro (1988)	Galician waters and Cantabrian Sea	4, 7, 9, and 12
Piñeiro and Pereiro (1993)	Galician waters and Cantabrian Sea	Hyaline band (7–10)

Courbin *et al.*, (2007) investigated the macrostructure pattern of hake otoliths from the Mediterranean Sea. Unsupervised statistical analysis of the distribution of the characteristics of these macrostructures led to the definition of three main macrostructure types: (i) a wide translucent zone close to the nucleus (WTZC), (ii) a wide translucent zone distant from the nucleus (WTZD), and (iii) a thin translucent zone or check (TTZ), associated with a large variation in distance to the nucleus. The TZZ type is therefore probably associated mainly with specific fish responses to environmental or endogenous factors. In contrast, the spatial extent of the WTZC and WTZD types are more clearly defined, suggesting that they are relevant at a population level, for instance in response to seasonal variations or major ontogenetic events.

The interpretation of the otolith growth pattern will be a fundamental step towards the establishment of validated age-reading criteria.

4.2 Lack of validated ageing criteria

Age validation is the confirmation of the temporal meaning of the growth structure used for age determination. However, it is not easy because the ageing has to be validated for the entire age range of the population and for spawning stocks in different geographic areas. Different techniques can be used for validation (Morales-Nin, 1997). Attempts were initially made using mark–recapture methods (Belloc, 1935; Fritz, 1959) for the study of European hake growth, but most researchers have relied on the interpretation of rings in bony structures (otoliths) and on analyses of length–frequency distributions, such as modal progression analysis and the separation of modal components (Gulland and Rosenberg, 1992). Although several techniques for making hake otoliths easier to examine have been employed (Christensen, 1964; Albrechtsen, 1968; Nichy, 1969), all researchers have encountered difficulties reaching a consistent interpretation of the pattern of growth ring (annual growth zone) formation on otoliths of this species.

The 1999 workshop permitted the establishment of age-reading criteria (Piñeiro and Saínza, 2003). The decrease in age-reading agreement that occurred at the last workshop, although detrimental, is obviously related to the use of hake estimation criteria that have not been validated. The use of validated criteria will encourage better results because it will reduce the subjectivity inherent in age reading, increase the confidence of readers in assigning a given age, and facilitate the identification of macrostructures in the otoliths that could easily be related to life-history events. The lack of knowledge of hake otolith growth patterns (see Section 4.1) makes it impossible to establish reliable ageing criteria.

4.3 Alternative methods

A variety of methods or approaches has been used in growth studies. The analyses of hake catch length distributions have been applied to hake age distribution (see Section 3.3). However, results of this methodology are difficult to interpret owing to the difficulty in splitting overlapping normal distributions or establishing the correspondence between normal distribution and year classes. Even where these analyses are possible, they are limited to the first age class or to the few age classes that make up the bulk of the landings.

Prior to 1997, the ICES Working Group for the Assessment of Southern Shelf Demersal Stocks (WGSSDS) – previously in charge of hake stock assessments – applied numerical methods (Kimura and Chikuni, 1987) to the annual length composition of catches in order to obtain a catch-at-age matrix for the assessment of the southern hake stock (ICES, 1993, 1994, 1995). However, the WGSSDS considered the use of ALKs to be more reliable for obtaining catch-at-age data (Piñeiro *et al.*, 2000). The application of length–frequency analysis is problematic because of the hake's extended spawning period. More than one mode may relate to the same year class, and the correct identification of length mode is subjective. Additional information can be achieved by age validation methods, including both direct and indirect approaches, in order to reduce growth and age uncertainties.

The mark–recapture of wild fish is one of the best methodologies to facilitate age-reading validation if the otoliths are chemically marked (with OTC) when the tagged fish are released. This approach, used successfully with hake, is very expensive because a large number of fish must be marked in order to obtain sufficient returns. Success is highly dependent on the recapture rate, which is often very low. The major difficulty is related to the tagging of small hake because of the high mortality rate that occurs during early life stages (Piñeiro *et al.*, 2007). For this reason, alternative approaches, such as otolith microstructure analysis, can be very useful in confirming growth in the first year of life.

Daily growth increment analysis is also a very important tool for the establishment of a criterion for the identification of the first annual ring. Furthermore, this analysis provides a better understanding of the typology of hake otolith macrostructure in relation to the biological meaning and mechanisms (endogenous and environmental) that control incremental deposition. The approach of comparing the structural pattern at micro- and macroscopic scales provides a new framework for the development of an alternative method of age estimation for this species.

Chemical-composition analysis of otolith growth marks may provide information about fundamental differences between opaque and translucent bands that will help to explain the processes governing their formation and, therefore, aid in their interpretation (Morales-Nin *et al.*, 2005; Tomás *et al.*, 2006). An electron probe study, based on calcium, strontium, and sodium, of some European hake populations has revealed relative changes in composition associated with opaque and translucent bands in their otoliths (Tomás *et al.*, 2006).

A combination of all methodologies, the description of otolith patterns, length–frequency analysis, daily growth increment analysis, and mark–recapture results is necessary to establish validated criteria for reliable age reading of hake otoliths. The application of each individual methodology in isolation, which has been the case until now, has not permitted the necessary advances in hake age reading.

4.4 Fast growth vs. slow growth

The debate about whether hake is a fast- or slow-growing species has continued since the 1930s (Hickling, 1933; Belloc, 1935). Results of recent mark–recapture studies demonstrate that growth is underestimated as a consequence of age overestimation (de Pontual *et al.*, 2006). Underestimation of growth during the first year of life has also been recently demonstrated, based on daily growth increments (Arneri and Morales-Nin, 2000; Morales-Nin and Moranta, 2004; Kacher and Amara, 2005; Belcari *et al.*, 2006; Piñeiro *et al.*, 2007). Bioenergetics studies also supported an underestimation of growth rate (Riis-Vestergaard *et al.*, 2000; see Section 3). The number of wide translucent zones (WTZ) detected by Courbin *et al.* (2007) were more in line with a fast-growth rather than a slow-growth hypothesis.

Scientists who provided age data for stock assessment purposes assumed a slow-growth hypothesis that is inherent to the standard criteria, which were agreed following the results of several workshops. However, complementary and recent studies indicate that the slow-growth hypothesis is less likely to be valid. Bearing in mind that the age criteria used are not validated and that the fast-growth hypothesis is consistent with mark–recapture results, a reliable and non-subjective methodology that allows accurate results is necessary. It is clear that conventional otolith age estimation needs to change in order to be consistent with the fast-growth hypothesis. According to the fast-growth hypothesis, hake at the end of the first, second, and third years of life would reach a TL of 25, 45, and 60 cm respectively, instead of 20, 29, and 37 cm (Piñeiro *et al.*, 2007), as estimated from otoliths using the internationally agreed ageing method (Piñeiro and Saínza, 2003).

5 Future work

5.1 Exchange programme

The objective of otolith exchanges is to estimate precision and relative/absolute bias in the age readings made by readers from different age-reading laboratories in order to determine whether or not these parameters are still within acceptable levels (ICES, 2008b). In this case, a new otolith exchange has been scheduled for 2009 to identify the nature of the translucent rings that contributed to the decrease in the precision of age estimations between the last two exchanges (2001 and 2003, where the CV increased from 25 to 48%) for fish of the same length range. Taking into account the known discrepancy between the standard ageing criteria and the results of mark–recapture experiments, the next exchange needs not only to assess the precision of age estimation but also to start developing new validated age-reading criteria. This can be achieved by using otoliths collected from both tagging material and previous workshop collections; it is essential to compile a collection of currently recognized known-age material that can be used for validation. The position of known annual translucent rings on otoliths from marked and recaptured fish can be compared with translucent rings identified by individual readers as annual rings on raster layers of images of the otoliths.

5.2 Otolith biometrics

The purpose of inferring age from the correlation between the age of a fish and otolith biometrics, such as otolith weight or two-dimensional otolith shape, is to estimate the proportions-at-age within a fish population rather than to assign ages to individual fish (FAbOSa, 2002; Francis and Campana, 2004). The development of an otolith growth model is hampered by the lack of validated material (mark–recapture or other data on known age); this prevents not only validation, but also the development of statistical methods to update historical data (ICES, 2006).

Recent advances in image acquisition and the development of image analysis techniques for fish otoliths offer new possibilities for the automation of aspects of image processing that can improve the objectivity of reader interpretation within a quality-control framework.

5.3 Otolith macrostructure typology

The low levels of precision in the age interpretation of older hake otoliths are partly the result of the complexity of the otolith macrostructure. Problems in age interpretation can only be resolved by establishing descriptors for the key features within the otolith macrostructure and developing a better understanding of their biological significance and the mechanisms that control the deposition of the growth rings. Courbin *et al.* (2007) have developed a methodological framework that, when applied to a set of Mediterranean hake otoliths, discriminates between two types of macrostructure that are clearly not random and are meaningful at a population or group level. However, the authors note that, although this proposed measure of similarity of otolith patterns can be applied to the patterns interpreted by experts with a view to quantifying interpretation differences, it would rely on a predetermined macrostructure typology agreed among experts.

5.4 Experiments in a controlled environment

Laboratory experiments to confirm the increment formation of known-age or chemically marked fish are common (Geffen, 1992). However, it is seldom possible to mimic in the laboratory natural conditions, such as photoperiodicity, temperature and feeding cycles, and vertical migrations. Nevertheless, it can be helpful to know when important events, such as growth and behaviour, occur at different stages in otolith growth pattern formation. This information can only be obtained from rearing experiments. Semi-intensive culture of the eggs of European hake has allowed descriptions to be made of larval development, growth during early life stages, and the influence of temperature on development and survival (Morales-Nin *et al.*, 2005; Bjelland and Skiftesvik, 2006). Studies on the correlation between otolith micro- and macrostructure and feeding, temperature, and fish growth require empirical observations in laboratory experiments. Recently, Martínez de Murguía *et al.* (2005) demonstrated that, with regular feeding, it is possible to keep adult hake in captivity for more than a year. This experiment opens the possibility of future research for studying the optical signal of otolith structures and other issues concerning their biology under controlled conditions.

5.5 Developing a new protocol for hake age estimation

The otolith exchange in 2009 will be followed by a Workshop on Age Estimation of European Hake (WKAEH), which will take place at IEO in Vigo, Spain, in October/November of the same year. Identification of the problematic translucent zones and a better understanding of the features that make them difficult to interpret are essential steps in the development of an improved protocol that can increase the precision of hake age estimation for stock assessments. A comparison of the position of known annual rings on otoliths from marked and recaptured fish, and the identification of these rings by experienced otolith readers during the exchange, will be a key topic of the workshop. Otolith microstructure analysis and observed growth rates from tagging experiments will be used to inform discussions on the improvement of the criteria required to distinguish annual growth rings from other rings, as will the results of recent research. The presence of stock assessment experts at this workshop, and their interaction with age readers, will help lead to the provision of age data that can be used reliably for stock assessments.

5.6 Summary of recommendations for future work

It is clear from the difficulties previously described that a great deal of work still needs to be done to develop stable and acceptable levels of precision for European hake age estimation. Recommendations include:

- an exchange programme (2009), using a set of otoliths (images) collected from both tagging material and previous workshop collections, for the purpose of intercalibration between age readers involved in stock assessment
- a workshop in 2009 to analyse and discuss the results of the exchange programme between age-reading laboratories
- a meeting between international experts on growth and age estimation and scientists involved in stock assessments, aimed at producing age data that can be reliably used for stock assessments

- provision of supplementary data in order to perform statistical approaches to estimating age distributions, including:
    - otolith biometrics and weight
    - historical data from length–frequency distribution from surveys and commercial catches
- continued collection of data and otolith for future work
- a large-scale tagging experiment covering both northern and southern stocks
- continuation of daily growth studies in order to establish seasonal growth variations and to locate the first annual ring by area and stock
- establishment of a typology of otolith macrostructure and an understanding of its biological meaning, as well as an understanding of the mechanisms (environmental and endogenous) that control otolith ring (growth zone) deposition
- development of a refined model of otolith formation, including growth patterns and otolith shape
- production of new validated standard criteria for age interpretation that take all the above items into account

Acknowledgements

We thank especially R. Millner for his help and valuable comments on earlier versions of this report. We also thank A. Leal and L. Rodriguez for their help with the bibliography and technical assistance.

6 References

Albrechtsen, K. 1968. A dyeing technique for otolith age reading. Journal du Conseil International pour l'Exploration de la Mer, 32: 278–280.

Alcázar, J., Carrasco, F., Llera, E., Menéndez, M., Ortea, J., and Vizcaíno, A. 1983. Recursos Pesqueros de Asturias. 3: Biología, Dinámica y Pesca de la Merluza en Asturias. Consejería de Agricultura y Pesca del Principado de Asturias, Gijón, Spain. 135 pp.

Aldebert, Y., and Recasens, L. 1995. Estimation de la croissance de merlu dans le Golfe de Lion par l'analyse des frequences de tailles. CIHEAM, Cahiers Options Méditerranéennes, Dynamique des Populations Marines, 10: 49–50.

Alemany, F., and Oliver, P. 1995. Growth of female hake in the Balearic Sea: a proposal of a new growth model with higher growth rates. Rapports et Procès-Verbaux des Réunions – Commission Internationale pour l'Exploration Scientifique de la Mer Méditerranée, 34: 236.

Alemany, F., Oliver, P., and Reñones, O. 1993. Growth of hake of Balearic Islands stock. *In* Etùde pour l'aménagement et la gestion des pêches en Méditerranée occidentale. Rapport final. CEE FAR. 474 pp.

Álvarez, P., and Cotano, U. 2005. Growth, mortality and hatch-date distributions of European hake larvae, *Merluccius merluccius* (L.), in the Bay of Biscay. Fisheries Research, 76: 379–391.

Álvarez, P., Fives, J., Motos, L., and Santos, M. 2004. Distribution and abundance of European hake *Merluccius merluccius* (L.), eggs and larvae in the North East Atlantic waters in 1995 and 1998 in relation to hydrographic conditions. Journal of Plankton Research, 26: 811–816.

Anon. 1998. Biological Studies of Demersal Fish to European Commission. Annex III of the Final Report to the Commission of European Communities (BIOSDEF, EU Study Contract 95/038). 522 pp.

Anon. 2000. Report of New Assessment and Biology of the Main Commercial Fish Species: Hake and Anglerfish of the Southern Shelf Demersal Stocks in the South Western Europe. Appendix I of the Final Report to the Commission of European Communities (DEMASSESS, EU Study Contract 97/015). 497 pp.

Arneri, E., and Morales-Nin, B. 2000. Aspects of the early life history of European hake from the central Adriatic. Journal of Fish Biology, 56: 1368–1380.

Bagenal, T. B. 1954. Growth rate of the hake (*Merluccius merluccius* L.) in the Clyde and other Scottish areas. Journal of the Marine Biology Association of the UK, 33: 69–95.

Beamish, R. J., and Fournier, D. A. 1981. A method for comparing the precision of a set of age determinations. Canadian Journal of Fisheries and Aquatic Sciences, 38: 982–983.

Belcari, P., Ligas, A., and Viva, C. 2006. Age determination and growth of juveniles of the European hake (*Merluccius merluccius* L., 1758). Fisheries Research, 78: 211–217.

Belloc, G. 1929. Étude monographique du merlu. Revue des Travaux de l'Office des Pêches Maritimes, Revue Trimestrielle, Tome II, Fasc. II (Paris). 67 pp.

Belloc, G. 1935. Etude monographique du merlu *Merluccius merluccius,* 3ème partie. Revue des Travaux de l'Office des Pêches Maritimes, 8: 145–202.

Bertignac, M., and de Pontual, H. 2007. Consequences of bias in age estimation on assessment of the northern stock of European hake (*Merluccius merluccius*) and on management advice. ICES Journal of Marine Science, 64: 981–988.

Bhattacharya, C. G. 1967. A simple method of resolution of a distribution into Gaussian components. Biometrics, 23: 115–135.

Birtwistle, W., and Lewis, H. M. 1925. Hake investigations. Reports of the Lancashire Sea Fisheries Laboratory, 1925: 36–56.

Bjelland, R. M., and Skiftesvik, A. B. 2006. Larval development in European hake (*Merluccius merluccius* L.) reared in a semi-intensive culture system. Aquaculture Research, 37: 1117–1129.

Campana, S. E. 2001. Accuracy, precision and quality control in age determination, including a review of the use and abuse of age validation methods. Journal of Fish Biology, 59: 197–242.

Casey, J., and Pereiro, F. J. 1995. European hake (*M. merluccius*) in the North-East Atlantic. *In* Hake: Fisheries, Ecology and Markets. Fish and Fisheries Series 15, pp. 125–147. Ed. by J. Alheit and T. J. Pitcher. Chapman and Hall, London. 508 pp.

Chang, W. Y. B. 1982. A statistical method for evaluating the reproducibility of age determination. Canadian Journal of Fisheries and Aquatic Sciences, 39: 1208–1210.

Christensen, J. M. 1964. Burning of otoliths, a technique for age determination of soles and other fish. Journal du Conseil Permanent International pour l'Exploration de la Mer, 29: 73–81.

Cimmaruta, R., Bondanelli, P., and Nascetti, G. 2005. Genetic structure and environmental heterogeneity in the European hake (*Merluccius merluccius*). Molecular Ecology, 14: 2577–2591.

Courbin, N., Fablet, R., Mellon, C., and de Pontual, H. 2007. Are hake otolith macrostructures randomly deposited? Insights from an unsupervised statistical and quantitative approach applied to Mediterranean hake otoliths. ICES Journal of Marine Science, 64: 1191–1201.

de Pontual, H., Bertignac, M., Battaglia, A., Bavouxet, G., Moguedet, P., and Groison, A. L. 2003. A pilot tagging experiment on European hake (*Merluccius merluccius*): methodology and preliminary results. ICES Journal of Marine Science, 60: 1318–1327.

de Pontual, H., Groison, A. L., Piñeiro, C., and Bertignac, M. 2006. Evidence of underestimation of European hake growth in the Bay of Biscay, and its relationship with bias in the agreed method of age estimation. ICES Journal of Marine Science, 63: 1674–1681.

Descamps, P., and Labastie, J. 1978. Note sur la lecture et l'interpretation des otolithes du merlu. ICES Document CM 1978/G:41. 21 pp.

Domínguez-Petit, R. 2007. Study of reproductive potential of *Merluccius merluccius* in the Galician Shelf. PhD thesis, University of Vigo. 253 pp.

Eltink, A. T. G. W. 1997. Horse mackerel otolith exchange in 1996. ICES Document CM 1997/H:24. 30 pp.

Eltink, A. T. G. W., Newton, A. W., Morgado, C., Santamaria, M. T. G., and Modin, J. 2000. Guidelines and tools for age reading comparisons, European Fish Ageing Network (EFAN), EFAN Report 3/2000. Available online at http://www.efan.no/NewsAndReports/EFANReport32000.htm.

FAbOSa. 2002. Final Report to the European Commission. EU Contract no. FAIR CT97 3402 for the FAbOSa (Fish Ageing by Otolith Shape Analysis) Project.

FAO. 1982. Report of the technical consultation on methodologies used for fish age-reading. FAO Fisheries Report 257. 109 pp.

Fariña, A. C., and Abaunza, P. 1991. Contribucion al estudio de los juveniles de merluza entre Cabo Villano y Cabo Prior (NW Galicia) mediante prospecciones pesqueras. Boletín del Instituto Español Oceanografía, 7: 155–163.

Fives, J. M., Acevedo, S., Lloves, M., Whitaker, A., Robinson, M., and King, P. A. 2001. The distribution and abundance of larval mackerel, *Scomber scombrus* L., horse mackerel, *Trachurus trachurus* L., hake *Merluccius merluccius* L., blue whiting *Micromesistus poutassou* (Risso,

1826) in the Celtic Sea and west of Ireland during the years 1986, 1989 and 1992. Fisheries Research, 50: 17–26.

Francis, R. I. C. C., and Campana, S. E. 2004. Inferring age from otolith measurements: a review and a new approach. Canadian Journal of Fisheries and Aquatic Sciences, 61: 1269–1284.

Fritz, R. L. 1959. Hake tagging in Europe and the United States, 1931–1958. Journal du Conseil Permanent International pour l'Exploration de la Mer, 24: 480–485.

García-Rodríguez, M., and Esteban, A. 1995. Algunos aspectos sobre la bilogía y pesca de la merluza mediterranea (*Merluccius merluccius* L.) en la Bahia de Santa Pola (Sureste de la Peninsula Iberica). Boletín. Instituto Español de Oceanografía, 11: 3–25.

García-Rodríguez, M., and Esteban, A. 2002. How fast does hake grow? A study on the Mediterranean hake (*Merluccius merluccius L.*) comparing whole otoliths readings and length frequency distributions data. Scientia Marina, 66: 145–156.

Geffen, A. J. 1992. Validation of otolith increment deposition rate. *In* Otolith Microstructure Examination and Analysis, pp. 101–113. Ed. by D. K. Stevenson, and S. E. Campana. Canadian Special Publication of Fisheries and Aquatic Sciences, 117. 126 pp.

Godinho, M. L., Afonso, M. H., and Morgado, C. 2001. Age and growth of hake *Merluccius merluccius* Linnaeus, 1758 from the Northeast Atlantic (ICES Division IXa). Boletín Instituto Español de Oceanografía, 17: 255–262.

Goñi, R. 1983. Growth studies of European hake (*Merluccius merluccius* L.) from the northwest African shelf. ICES Document CM 1983/G:10. 3 pp.

Goñi, R., and Piñeiro, C. 1988. Study of the growth pattern of European hake (*Merluccius merluccius* L.) from the southern stock: ICES Divisions VIIIc and IXa. ICES Document CM 1988/G:18. 13 pp.

Guichet, R. 1988. Étude de la croissance du merlu Européen (*Merluccius merluccius* L.) au cours de ses premières années. ICES Document CM 1988/G:53. 27 pp.

Guichet, R., Quero, J. C., and Labastie, J. 1973. Estimation de la composition du stock du merlu au nord et à l'ouest de l'Irlande. ICES Document CM 1973/G:5. 9 pp.

Guichet, R., Quero, J. C., and Labastie, J. 1974. Composition du stock du merlu au nord et a l'ouest de l'Irlande (deuxiéme note). ICES Document CM 1974/G:8. 5 pp.

Gulland, J., and Rosenberg, A. 1992. A review of length-based approaches to assessing fish stocks. FAO Technical Report 323. 100 pp.

Hickling, C. F. 1933. The natural history of the hake. Part 3. Seasonal changes in the condition of hake. UK Fishery Investigations Series II, 12(1). 120 pp.

ICES. 1979. Report of the Working Group on Assessment of the Hake Stocks. 28–31 May 1979, Charlottenlund. ICES Document CM 1979/G:27. 65 pp.

ICES. 1983. Confrontation Technique sur les Methodologies Utilisées pour la Lecture des Otolithes de Merlu. *In* Report of the Working Group on Assessment of Hake Stocks, pp. 49-55. 29 June–6 July 1983, Copenhagen. ICES Document CM 1983/Assess:20. 55 pp.

ICES. 1984. Report of the *ad hoc* Hake Otolith Working Group, 28 May–1 June 1984, Lowestoft. ICES Document CM 1984/G:74. 19 pp.

ICES. 1986. Report of the *ad hoc* Study Group on Hake, 3–6 June 1986, Lowestoft. ICES Document CM 1986/G:93. 19 pp.

ICES. 1991. Report of the Working Group on the Assessment of the Stocks of Hake. 29 April–8 May 1991. ICES Document CM 1991/Assess:20. 25 pp.

ICES. 1993. Report of the Working Group on the Assessment of Southern Shelf Demersal Stocks, Copenhagen, 8–17 September 1992. ICES Document CM 1993/Assess:3. 181 pp.

ICES. 1994a. Report of the Working Group on the Assessment of Southern Shelf Demersal Stocks, Copenhagen, 6–15 September 1993. ICES Document CM 1994/Assess:3. 92 pp.

ICES. 1994b. Report of the Workshop on Sampling Strategies for Age and Maturity, Copenhagen, 3–9 February 1994. ICES Document CM 1994/D:1. 67 pp.

ICES. 1995. Report of the Working Group on the Assessment of Southern Shelf Demersal Stocks, Copenhagen, 31 August–6 September 1994. ICES Document CM 1995/Assess:6. 598 pp.

ICES. 2000. Report of the Working Group on the Assessment of Southern Shelf Demersal Stocks, Copenhagen, 14–23 September 1999. ICES Document CM 2000/ACFM:4. 744 pp.

ICES. 2003. Report of the Working Group on the Assessment of Southern Stock of Hake, Monk, and Megrim (WGHMM), Lisbon, Portugal, 21–30 May 2002. ICES Document CM 2003/ACFM:01. 499 pp.

ICES. 2006. Report of the Study Group on Ageing Issues of Baltic Cod (SGABC), 16–19 May 2006, Gdynia, Poland. ICES Document CM 2006/BCC:08. 45 pp.

ICES. 2007a. Report of the Working Group on the Assessment of Southern Shelf Stocks of Hake, Monk, and Megrim (WGHMM), 8–17 May 2007, Vigo, Spain. ICES Document CM 2007/ACFM:21. 700 pp.

ICES. 2007b. Report of the ICES Advisory Committee on Fisheries Management, Advisory Committee on the Marine Environmental and Advisory Committee on Ecosystems, 2007. Book 7: Bay of Biscay and Western Iberia. ICES Advice. 94 pp.

ICES. 2008a. Report of the Working Group on the Assessment of Southern Shelf Stocks of Hake, Monk, and Megrim (WGHMM), 30 April– 6 May, Copenhagen. ICES Document CM 2008/ACOM:07. 622 pp.

ICES. 2008b. Report of the Planning Group on Commercial Catches, Discards and Biological Sampling (PGCCDBS), 3–7 March 2008, Cyprus, pp. 70–75. ICES Document CM 2008/ACOM:29. 91 pp.

Iglesias, S., and Dery, L. 1981. Age and growth studies of hake (*Merluccius merluccius,* L.) from ICES Divisions VIIIc and IXa. ICES Document CM 1981/G:38. 6 pp.

Kacher, M., and Amara, R. 2005. Distribution and growth of 0-group European hake in the Bay of Biscay and Celtic Sea: a spatial and interannual analysis. Fisheries Research, 71: 373–378.

Kimura, D. K., and Chikuni, S. 1987. Mixtures of the empirical distributions: an iterative application of the age–length key. Biometrics, 13: 23–35.

Lannin, A. 2006. The biology, dynamics and fisheries for hake (*Merluccius merluccius*) in the waters around Ireland. PhD thesis, University College Cork. 182 pp.

Lucio, P., Murúa, H., and Santurtún, M. 2000. Growth and reproduction of hake (*Merluccius merluccius*) in the Bay of Biscay during the period 1996–1997. Ozeanografika, 3: 325–354.

MacDonald, P., and Pitcher, T. 1979. Age groups from size frequency data: a versatile and efficient method of analysing distribution mixtures. Journal of the Fisheries Research Board of Canada, 36: 987–1001.

Martínez de Murguía, A., Leé Herrero, D., Gilisagasti, J., Sanchez, S., Casares, P., and Lucio, P. 2005. First attempt at keeping *Merluccius merluccius* European hake in closed tanks. *In* Proceedings of the 21st Meeting of the European Union of Aquarium Curators (EUAC), Vienna, Austria. Bulletin de l'Institut Océanographique, Monaco, 77: 133–139.

Meriel-Bussy, M. 1966. La croissance du merlu dans le golfe de Gascogne. ICES Document CM 1966/G:17. 5 pp.

Morales-Nin, B. 1997. Review on the European hake age determination and validation problems. European Fish Ageing Network, EFAN Report, 1/1997.

Morales-Nin, B., and Aldebert, Y. 1997. Growth of juvenile *Merluccius merluccius* in the Gulf of Lions (NW Mediterranean) based on otolith microstructure and length–frequency analysis. Fisheries Research, 30: 77–85.

Morales-Nin, B., and Moranta, J. 2004. Recruitment and post-settlement growth of juvenile *Merluccius merluccius* on the western Mediterranean shelf. Scientia Marina, 63: 399–409.

Morales-Nin, B., Torres, G. J., Lombarte, A., and Recasens, L. 1998. Otolith growth and age estimation in the European hake. Journal of Fish Biology, 53: 1155–1168.

Morales-Nin, B., Bjelland, R. M., and Moksness, E. 2005. Otolith microstructure of a hatchery reared European hake (*Merluccius merluccius*). Fisheries Research, 74, 300–305.

Murúa, H. 2006. Reproductive fundamentals for the estimation of egg production of the European hake, *Merluccius Merluccius*, in the Bay of Biscay. PhD thesis, University of Basque Country (Spain). 158 pp.

Murúa, H., and Motos, L. 2006. Reproductive strategy and spawning activity of the European hake (*Merluccius merluccius* L.) in the Bay of Biscay. Journal of Fish Biology, 69: 1288–1303.

Murúa, H., Motos, L., and Marrale, D. 1996. Reproductive modality and batch fecundity of the European hake, *Merluccius merluccius*. ICES Document CM 1996/G: 40. 28 pp.

Murúa, H., Lucio, P., Santurtún, M., and Motos, L. 2006. Seasonal variation in egg production and batch fecundity of European hake *Merluccius merluccius* (L.) in the Bay of Biscay. Journal of Fish Biology, 69: 1304–1316.

Mytilineou, Ch., and Vassilopoulou, V. 1988. The reproductive cycle and sex ratio of hake, *Meluccius merluccius*, in Patraikos and Korinthiakos gulfs and the Ionian Sea. Proceedings of the 4th Hellenic Congress of Ichthyologists, 4: 164–177.

Nichy, F. E. 1969. Growth patterns on otoliths from young silver hake, *Merluccius bilinearis* (Mitch). International Commission for the Northwest Atlantic Fisheries Research Bulletin, 6: 107–117.

Oliver, P. 1991. Dinámica de la población de Merluza (*Merluccius merluccius*, L.) de Mallorca. Reclutamiento, crecimiento y mortalidad. PhD thesis, Universitat de les Illes Baleares. 392 pp.

Oliver, P., Alvarez, F., and Morales-Nin, B. 1989. Report of the age-reading workshops on Mediterranean European hake and sardine, 10–15 April 1989, Palma de Mallorca, Spain. FAO Rapport sur les Pêches, 447, 79–86.

Orsi-Relini, L., Cappanera, M., and Fiorentino, F. 1989. Spatial-temporal distribution and growth of *Merluccius merluccius* recruits in the Ligurian Sea: observations on the 0 group. Cybium, 13: 263–270.

Palomera, I., Olivar, M. P., and Morales-Nin, B. 2005. Larval development and growth of the European hake *Merluccius merluccius* in the northwestern Mediterranean. Scientia Marina, 69: 251–258.

Pannela, G. 1971. Fish otoliths: daily growth layers and periodical patterns. Science, 173: 1124–1127.

Pérez, N., and Pereiro, F. J. 1985. Aspectos de la reproducción de la merluza (*Merluccius merluccius*) de la plataforma gallega y cantábrica. Boletín del Instituto Español Oceanografía, 2: 39–47.

Pérez, N., Pereda, P., Uriarte, A., Trujillo, V., Olaso, S., and Lens, S. 1996. Descartes de la flota española en el área del ICES. Datos y Resúmenes Instituto Español de Oceanografía, 2. 142 pp.

Petersen, C. G. 1891. Eine Methode zur Bestimmung des Alters und Wuchses der Fische. Mitteilungen. Deutscher Seefisherei-Verein, 11: 226–235.

Piñeiro, C. 2000. Report on workshop on hake otolith age reading, June 1997, Vigo. European Fish Ageing Network, EFAN Report, 6/2000.

Piñeiro, C., and Hunt, J. J. 1989. Comparative study on growth of European hake (*Merluccius merluccius* L.) from southern stock using whole and sectioned otoliths, and length frequency distributions. ICES Document CM 1989/G:37. 16 pp.

Piñeiro, C., and Meixide, M. 1995. Analysis of results obtained in the Spanish-French meeting about otolith age reading of European hake. *In* Report of the Working Group on the Assessment of Southern Shelf Demersal Stocks, Copenhagen, 6–15 September 1994. ICES Document CM 1995/Assess:6. 598 pp.

Piñeiro, C. G., and Pereiro, J. A. 1993. Study of the growth pattern of juvenile European hake (*Merluccius merluccius* L.) using the whole otoliths and length frequency from commercial catches and groundfish surveys. ICES Document CM 1993/G:12. 14 pp.

Piñeiro, C., and Saínza, M. 2002. Report of 3rd exchange on European hake age readings. Annex of the Final Report of EU Contract 99/009, Improving Sampling of Western and Southern European Atlantic Fisheries – SAMFISH.

Piñeiro, C., and Saínza, M. 2003. Age estimation, growth and maturity of the European hake, *Merluccius merluccius* (Linnaeus, 1758) from Iberian Atlantic waters. ICES Journal of Marine Science, 60: 1086–1102.

Piñeiro, C. G., Fariña, A. C., and Abaunza, P. 1992. Resultados de las campañas realizadas en las zonas de veda al arrastre en la plataforma continental Gallega (NW España), 1989–1990. Informes Técnicos Instituto Español Oceanografía, 130. 39 pp.

Piñeiro, C., Afonso, M. H., Arego, S., Bellail, R., Labastie, J., Loureiro, I., Lucio, P., *et al.* 2000. Report of 2nd international workshop on hake otolith age reading, February 1999, Vigo, Spain. European Fish Ageing Network, EFAN Report, 7/2000.

Piñeiro, C., Saínza, M., Morgado, C., de Pontual, H., Hoey, S., Beattie, S., Lucio, P., *et al.* 2004. Report of 4th Exchange on European Hake Age Readings, 16–20 October 2004, Vigo, Spain. ICES Document CM 2005/ACFM:02. 18 pp.

Piñeiro, C., Rey, J., de Pontual, H., and Goñi, R. 2007. Tag and recapture of European hake (*Merluccius merluccius* L.) off the northwest Iberian Peninsula: first results support fast-growth hypothesis. Fisheries Research, 88: 150–154.

Piñeiro, C., Rey, J., de Pontual, H., and García, A. 2008. Growth of Northwest Iberian juvenile hake estimated by combining sagittal and transversal otolith microstructure analyses. Fisheries Research, 93: 173–178.

Recasens, L. 1992. Dinámica de poblaciones i pesqueria del lluc (*Merluccius merluccius*) al Golfo de Lleon i la mar Catalana. PhD thesis, Universitat de Barcelona. 398 pp.

Riis-Vestergaard, J., Velasco, F., Hill, L., and Olaso, I. 2000. Food consumption of European hake (*Merluccius merluccius*) estimated by application of a bioenergetics model: is the growth of hake underestimated? ICES Document CM 2000/Q:12. 12 pp.

Robles, R., Pereiro, J. A., Iglesias, S., and Pereiro, F. J. 1975. Étude sur la croissance du merlu Européen, *Merluccius merluccius* L. du NW de L'Espagne. ICES Document CM 1975/G:8. 17 pp.

Silva, L., Vila, Y., Acosta, J. J., and Tornero, J. 2007. Size at first maturity in European hake: estimates based on different maturity criteria. Working Document (WD10) presented to the Workshop on Sexual Maturity Staging of Hake and Monk (WKMSHM), 21–24 November 2007, Lisbon, Portugal. ICES Document CM 2007/ACFM:34. 82 pp.

Tomás, J., Geffen, A. J., Millner, R. S., Piñeiro, C., and Tserpes, G. 2006. Elemental composition of otolith growth marks in three geographically separated populations of European hake (*Merluccius merluccius*). Marine Biology, 148: 1399–1413.

Velasco, F. 2007. Alimentación de la merluza europea (*Merluccius merluccius* L.) en el mar Cantábrico. PhD thesis, Complutense University of Madrid. 257 pp.

Velasco, F., and Olaso, I. 1998. European hake (*Merluccius merluccius* L. 1758) feeding in the Cantabrian Sea: seasonal, bathymetric and length variations. Fisheries Research, 38: 33–44.

Velasco, F., Olaso, I., and Sánchez, F. 2003. Annual variations in the prey of demersal fish in the Cantabrian Sea and their implications for foodweb dynamics. ICES Marine Science Symposia, 219: 408–410.

von Bertalanffy, L. 1938. A quantitative theory of organic growth. Human Biology, 10: 181–213.

Zar, J. H. 1996. Biostatistical Analysis, 3rd edn. Prentice-Hall, Englewood Cliffs, NJ. 662 pp.

Acronyms

ALK	age–length key
APE	average percentage error
AZTI–Tecnalia	Instituto Tecnológico, Pesquero y Alimentario (Technological Center, Fishing and Food Research)
BIOSDEF	Biological Studies of Demersal Fish to the European Commission (EU Study Contract 95/038)
CV	coefficient of variation
DEMASSESS	New Assessment and Biology of the Main Commercial Fish Species: Hake and Anglerfish of the Southern Shelf Demersal Stocks in the South Western Europe (EU Study Contract 97/015)
IEO	Instituto Español de Oceanografía (Spanish Institute of Oceanography)
IFREMER	Institut Français de Recherche pour l'Exploitation de la Mer (French Research Institute for the Exploitation of the Sea)
WGHMM	Working Group on the Assessment of Southern Shelf Stocks of Hake, Monk, and Megrim

Titles published in the ICES Cooperative Research Report series

No.	Title	Price (Danish Kroner)
293	The effect of climate change on the distribution and abundance of marine species in the OSPAR Maritime Area. 46 pp.	170
292	ICES Zooplankton Status Report 2006/2007. 164 pp.	150
291	ICES Report on Ocean Climate 2007. 60 pp.	30
290	Changes in surface CO_2 and ocean pH in ICES shelf sea ecosystems. 35 pp.	70
289	ICES Report on Ocean Climate. 56 pp.	70
288	Structure and dynamics of the North Sea benthos. 258 pp.	350
287	Collection of acoustic data from fishing vessels. 83 pp.	60
286	Acoustic seabed classification of marine physical and biological landscapes. 183 pp.	130
285	Results of the spring 2004 North Sea ichthyoplankton surveys. 59 pp.	60
284	Status of introductions of non-indigenous marine species to the North Atlantic and adjacent waters 1992–2002. 149 pp.	80
283	Alien Species Alert: *Undaria pinnatifida* (wakame or Japanese kelp). 36 pp.	60
282	Incorporation of process information into stock–recruitment models. 152 pp.	90
281	Zooplankton Monitoring Results in the ICES Area: Summary Status Report 2004/2005. 43 pp.	90
280	ICES Report on Ocean Climate. 47 pp.	70
279	Protocol for the Use of an Objective Mesh Gauge for Scientific Purposes. 8 pp.	40
278	Description of the ICES HAC Standard Data Exchange Format, Version 1.60. 86 pp.	60
277	The intentional introduction of the marine red king crab Paralithodes camtschaticus into the Southern Barents Sea. 18 pp.	60
276	Zooplankton Monitoring Results in the ICES Area: Summary Status Report 2003/2004. 34 pp.	80
275	The Annual ICES Ocean Climate Status Summary 2004/2005. 2005. 37 pp.	80
274	Spawning and life history information for North Atlantic cod stocks. 152 pp.	90
273	Guidance on the Application of the Ecosystem Approach to Management of Human Activities in the European Marine Environment. 22 pp.	40
272	Ecosystem Effects of Fishing: Impacts, Metrics and Management Strategies. 177 pp.	70
271	Vector Pathways and the Spread of Exotic Species in the Sea. 25 pp.	60
270	The *Nephrops* fisheries of the Northeast Atlantic and Mediterranean – A review and assessment of fishing gear design. 38 pp.	50
269	The Annual ICES Ocean Climate Status Summary 2003/2004. 32 pp.	60
268	The DEPM Estimation of Spawning-Stock Biomass for Sardine and Anchovy. 87 pp.	90
267	Report of the Thirteenth ICES Dialogue Meeting: Advancing scientific advice for an ecosystem approach to management: collaboration among managers, scientists, and other stakeholders. 59 pp.	50
266	Mesh Size Measurement Revisited. 56 pp.	80
265	Trends in important diseases affecting the culture of fish and molluscs in the ICES area 1998–2002. 26 pp.	40

These publications can be ordered from ICES Secretariat, H. C. Andersens Boulevard 44–46, DK-1553 Copenhagen V, Denmark, fax: +45 33 93 42 15, e-mail: info@ices.dk. An invoice including the cost of postage and handling will be sent. Publications are usually dispatched within one week of receipt of payment. Further information about ICES publications, including ordering and payment by credit card, cheque, and bank transfer, can be found at www.ices.dk under "Publications".