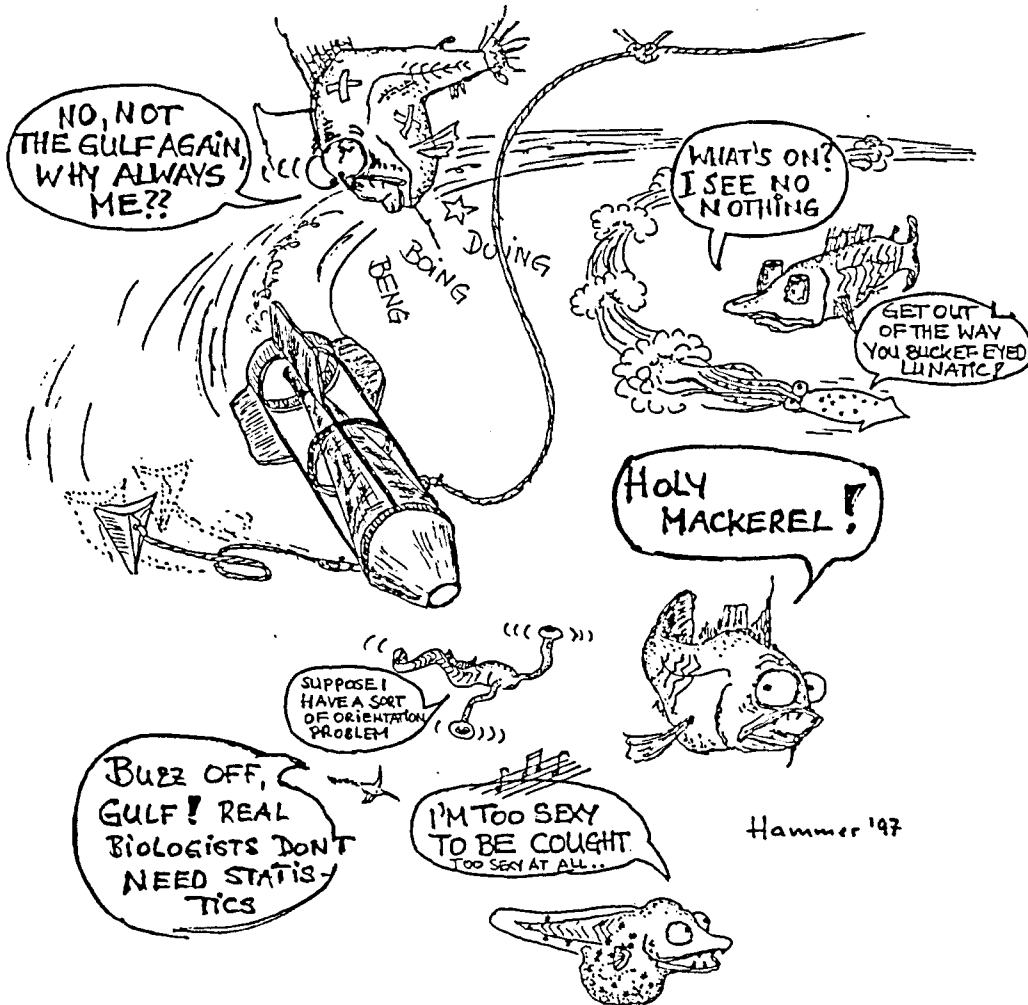# REPORT OF THE

WORKING GROUP ON MACKEREL AND HORSE MACKEREL EGG SURVEYS

Lisbon, Portugal
3–7 February 1997

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International Council for the Exploration of the Sea

Conseil International pour l'Exploration de la Mer

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1 INTRODUCTION

1.1 Terms of Reference

At the ICES Annual Science Conference in Reykjavík, Iceland in October 1996 it was decided that (C.Res.1996/2:36) the Working Group on Mackerel and Horse Mackerel Egg Surveys [WGMEGS] (Chairman: Mr J.H. Nichols, UK) would meet in Lisbon, Portugal from 3–7 February 1997 to:

a) co-ordinate the timing and planning of the 1998 Mackerel/Horse Mackerel Egg Surveys in ICES Sub-areas VI to IX for estimating spawning stock size;

b) further evaluate the use of Generalised Additive Modelling in survey planning and analysis of egg survey data with reference to the results of the analysis of the 1989, 1992 and 1995 surveys and the comments of the 1996 Working Group on Mackerel, Horse Mackerel, Sardine and Anchovy (WGMHSA);

c) review all the fecundity and atresia data collected in the western and southern areas for mackerel with particular reference to the significance of any inter-annual differences in the values measured. Advise the MHSA Working Group on any changes which should be made to the values of fecundity and atresia used by them in their analysis of the 1995 egg survey data;

d) co-ordinate the planning of sampling for maturity of both mackerel and horse mackerel to be used for histological analysis;

e) examine the basis for the different mackerel maturity ogive used in 1986. Estimate appropriate maturity ogives from the survey data for use in the calculation of SSB in 1992 and 1995 with an estimate of the CV;

f) examine ways of combining the mackerel egg survey data for the western and southern areas to produce a single estimate of egg production for the combined North East Atlantic Mackerel;

g) consider any advice from the Plankton Sampler Study Group on standardising plankton sampling gear for the 1998 egg surveys.

1.2 Participation

The Working Group met in Lisbon (Portugal) from 3–7 February 1997 with the following participants:

Borges, Fatima	Portugal
Eltink, Guus	Netherlands
Farinha, Anabela	Portugal
Hammer, Cornelius	Germany
Iversen, Svein	Norway
Martins, M. Manuel	Portugal
Molloy, John	Ireland
Motos, Lorenzo	Spain
Nichols, John*	UK (England & Wales)
Porteiro, Carmela	Spain
Reid, Dave	UK (Scotland)
Sola, Amor	Spain
Witthames, Peter	UK (England & Wales)

*Chairman

The following attended as observers:

Costa, Ana Maria	Portugal
Cunha, Maria Emilia	Portugal
Leopold, Mardik	Netherlands
Meneses, Isabel	Portugal
Santos, Miguel	Portugal

2   BIOLOGICAL ASPECTS

2.1   Egg Staging and Exchange Programme

A comparison of staging of mackerel eggs was carried out in conjunction with the 1995 surveys (Anon., 1996). The results showed a reasonable consistency between countries for total stage 1 eggs although there were some problems in differentiating between stages 1A and 1B and between stages 2 and 3.

It was reported that some countries experienced difficulties in distinguishing between mackerel and horse mackerel eggs in some of their survey samples (Anon., 1996).

It is recommended that a further exchange of egg samples, for comparison of staging between participants, is carried out in conjunction with the 1998 egg surveys. The egg samples for this experiment should include mixed eggs of both species in all stages of development.

The egg exchange programme will be co-ordinated by S.P. Milligan, CEFAS, Lowestoft Laboratory, UK. and could be started before the 1998 surveys begin.

2.2   Egg Stage Duration

No further observations have been made on the rate of development of either mackerel or horse mackerel eggs since those reported on in 1994 (Anon., 1994). The relationships between temperature and rate of development of stage 1 mackerel and horse mackerel eggs, to be used for the calculation of daily egg production in the 1998 surveys, are given in Section 6.4.7.

2.3   Exchange Programme for Residual Fecundity and Atresia Estimation

A comparison of atresia estimates made by CEFAS Lowestoft and Aberdeen University (subcontracted to SOAEFD) presented to the Working Group showed significant differences in the estimation of atresia intensity (see Section 2.5). The Working Group recommends that the exchange of histological atresia slides takes place between institutes (CEFAS Lowestoft, Aberdeen University, IEO, IPIMAR for mackerel and RIVO-DLO, IEO, IPIMAR and FRC for horse mackerel). The exchange will be coordinated by Mr Witthames (CEFAS) for mackerel and Mr Vingerhoed (RIVO-DLO) for horse mackerel and will start in January 1998. For this exchange, slides previously analysed from the 1995 surveys (including the fish weight and ovary volume data), should be used to complete the analysis. In the analysis both residual fecundity and atresia should be considered. If differences are found a series of atresia pictures and/or fields analysed should be circulated by the co-ordinators and the process repeated.

2.4   Application of the Surveys to Other Species

2.4.1   Ichthyoplankton

The ICES triennial egg surveys, although primarily focused on mackerel and horse mackerel, provide a unique opportunity to collect information on the abundance and distribution of other fish populations. For this to be true, the potential target populations should produce pelagic eggs within the spatio-temporal sampling boundaries of the survey, i.e. the European continental shelf and outer approaches from the Iberian Peninsula to West of Scotland from January-February to July.

Some countries have previously shown interest in re-analysing the plankton samples after the primary processing for mackerel and horse mackerel eggs. Different papers have been published describing the larval communities found in the surveys in particular years (Horstman and Fives, 1994; O'Brien and Fives, 1995). The distribution and abundance of Bay of Biscay anchovy eggs was studied by Santiago and Eltink (1988). More recently, the information contained in the samples regarding the spatio-temporal patterns of abundance of eggs and larvae of mackerel, horse mackerel, blue whiting and hake, has been re-visited in the framework of the EU supported SEFOS project. An invaluable set of data has been gathered and the results, described in the final report of the SEFOS project, provided a much better understanding of the spatio-temporal patterns of distribution of the target species of eggs and larvae in relation to environmental conditions.

Concerning the potential assessment of species other than mackerel and horse mackerel, by using the results of the cruises, a working document was presented showing the potential application of this technique to the European hake population (Motos et al., WD1997). The document stated that the triennial egg survey covers a major part of the spawning areas and periods for the European hake. Hake has an extensive spawning area, this being along the shelf-edge and outer shelf region from the Iberian Peninsula and Biscay to northern Scotland and southern Norway. Spawning starts in the south in winter and finishes at the northern end of the distribution at around July-August. Spawning seems to start on the shelf edge and moves on to the shelf as the season progresses. The egg production records from the ICES triennial surveys can be used as abundance indices of the spawning population. When accompanied with unbiased fecundity estimations, absolute estimates of the spawning stock biomass may be attainable.

The document discussed the possibilities of the application of ichthyoplankton methods for the assessment of the European hake population. The use of the surface adhesion test (Porebski, 1977) has proven efficient in eliminating any uncertainty in egg identification. Following the standard techniques used in the traditional mackerel/horse mackerel surveys, estimates of daily egg production for hake were derived from a series of egg cruises carried out in Divisions VIIIa,b during the 1995 spawning season. The values of daily egg production for the entire area at peak spawning were quantified, together with available daily fecundity figures (Murua et al., 1996), to give estimates of spawning biomass for the sampled area. The results obtained for the 1995 spawning season were compared with the results of egg production obtained from a data set collected at a similar period in 1983.

One of the main problems found in the study was the low density of hake eggs generally found in the field, peak values ranging from 100 to 200 eggs per m^2. Nevertheless the assessment of hake egg abundance is considered to be tractable provided that the volume of water sampled is large enough (Motos et al., WD97). The authors concluded that 50 m^3 of sea water is the minimum volume to be filtered, in a standard plankton tow, in order to quantify hake egg abundance.

The Working Group agreed in principle that samples can be made available on request to the Institutes interested in further processing them. The Working Group recommends that all fish eggs be sorted from the samples collected in 1998. The Institutes taking part in the surveys, who have an interest in further processing the samples will take the necessary action. The remaining institutes should make the samples available to other Institutes in the following ways. Whenever practical, sardine and hake eggs should be identified and sent in separate vials to C. Porteiro (IEO Vigo) and to L. Motos (AZTI, San Sebastián), respectively. Sorted but unidentified eggs should be sent to L. Motos (AZTI, San Sebastián) in separate vials. Unsorted samples (other than for mackerel and horse mackerel eggs) collected north of 48° should be sent to J. Fives (University College of Galway) for further processing of fish larvae and zooplankters, whereas samples collected South of 48° should be sent to L. Motos (AZTI, San Sebastián). Samples collected south of 43° should be sent to F. Borges (IPIMAR, Lisbon). J. Fives and L. Motos should co-ordinate all the actions necessary to eventually get the largest amount of information extracted from the 1998 egg survey samples. It would be desirable that these studies result in collaborative technical and scientific papers from the Institutes involved in the surveys and those further processing the samples.

2.4.2   Observations of cetaceans and seabirds

During the 1995 mackerel/horse mackerel survey a study was carried out on board of the Dutch vessel, RV "Tridens", that looked into the possibility of using this survey for obtaining data on cetacean and seabird distribution and abundance. The egg surveys are a very useful platform for studying both cetaceans (Leopold & Couperus, 1995) and seabirds (van der Meer & Leopold, 1995).

The methods and results of the cetacean work during the 1995 pilot study were presented to the working group and copies of the report to the EU were handed out. Support was sought for cetacean/seabird workers to join the vessels involved in the 1998 survey. Ideally, each vessel should have three platforms, each manned by three observers. In reality, this is only possible on "Tridens". The Norwegian vessel may be able to take up to six observers on board. Most other vessels have room for a maximum of two extra people, and can only be used for seabird counts or for uncorrected cetacean observations. Such uncorrected observations will be useful, especially if these can be corrected by using results from the two ships where at least two platforms can be used. Furthermore, results collected from the smaller ships for cetaceans that are highly visible, like the large whales will be useful.

Targeted scientific questions include:

1. Assessment of distribution and numbers of fin and pilot whales off western Iberia during January-March

2. Assessment of distribution and numbers of seabirds off western Iberia during January-March

3. Assessment of distribution and numbers of whales, dolphins and seabirds in the Bay of Biscay in spring to early summer (March-July)

4. Assessment of distribution and numbers of the stock of White-sided Dolphins in the Bay of Biscay and waters west of Ireland and Scotland, noting that this species is of concern as a by-catch species in this area, but that its (local) population size has never been estimated.

The possibilities of using the 1998 egg surveys for these studies should be pursued by Mardik Leopold (Netherlands).

2.5   Review of Mackerel Fecundity and Atresia

The ICES Working Group on the Assessment of Mackerel, Horse Mackerel, Sardine and Anchovy requested a review of all the fecundity and atresia data collected in the western and southern areas with particular reference to the significance of any inter-annual changes in the values measured. It also required advice on the values of fecundity and atresia to use in the 1995 egg survey assessment. A working document was prepared by Witthames and Maxwell (WD 1997) in response to the above which is included in this report as Appendix 1.

The realised fecundity (F_{real}) is derived by subtracting atresia (F_{atr} loss of developing eggs per gram female from the ovary during spawning) from the relative potential annual fecundity (F_{pot} eggs per gram female). F_{real} is the denominator to the annual egg production (Egg_{annual}) in the equation to estimate female spawning biomass (B):

$$B = \frac{Egg_{annual}}{F_{real}}$$

F_{atr} is estimated in the population using the following equation:

$$F_{atr} = I_{int} \times Pr_{ev} \times \frac{D}{S}$$

where; I_{atr} = mean number of dead eggs per fish per gram total weight but excluding fish with no atresia present, P_{rev} = proportion of fish with atresia. S = the duration of spawning estimated as 60 days (Dawson, 1986; Eltink, 1987) D = the duration of the atretic stage estimated as 7.5 days (Anon., 1993). Previous results and development of the methods were described and reviewed in Anon., 1990, 1993, 1996. This review uses all the available data except the fecundity estimates prior to 1989 (1977 and 1986) because these surveys did not quantify atresia.

Review of Methods and Sampling to Estimate Atresia

The majority of fish were found to contain no atretic oocytes and in the remainder I_{atr} has a log normal distribution. Because of the large number of zero values there is not a suitable transformation for the whole data set. The previous approach using geometric means to estimate mean I_{atr} and inclusion of all zero values in the P_{rev} parameter was adopted for this study.

Samples were selected from the 1995 atresia data to test for variation that was attributable to laboratory methodology independent of biological factors. The selection was based on identifying fish sampled serially (n=155) from the various trawl hauls made in 1995 which were alternately distributed to the participating laboratories (Aberdeen University subcontracted to SOAEFD and CEFAS, Lowestoft). Table 2 in the appendix shows that there was no significant difference in the estimation of P_{rev} (p=0.958) which was 31% and 29% for Aberdeen University and CEFAS respectively. However, this was not the case for I_{atr} where the Aberdeen results were 2.6 times higher and significantly different (appendix Tables 3 & 4 P=0.0012 n =47) to CEFAS. Because this large variance was found in only a small part of the total analysis its effect on F_{real} would carry less weight. The Egg Production Working Group requested further work to remove this source of variance to improve the overall precision of atresia estimation (see Section 2.3).

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Tables 5 and 6 in the appendix show that the sampling levels achieved in the three survey years was very unequal both by station, and by month. Cruise leaders must try to ensure a uniform distribution of fish sampling for each cruise as specified in section 6.5.2 to improve the estimate of atresia within the overall constraint of successfully carrying out the egg survey.

Annual Variation in F_{pot} in 1989, 1992 and 1995 in the Western Area

Tables 7 and 8 in the appendix show that fecundity in the Western area declined significantly with a year effect as either a class variable (p=0.047) or as a continuous variable showing a linear trend (P=0.013). The estimated values of F_{pot} in Table 8 were 1543 se. 31, 1485 se. 33 and 1437 se. 29 in 1989, 1992 and 1995 respectively. They are different to those used in the biomass calculation because they are sample means and deviate from the population means if the relationship between F_{pot} and fish weight is significant, as it was in 1992 (Anon., 1993).

Variation of F_{pot} in the Western and Southern Areas

The relationship between F_{pot} and fish weight in the Southern and Western areas during 1995 was very different (appendix Figure 1 and Tables 9–10) but the cause was found to lie in the fish weight variable and not the area class variable (P>.05). A weak positive relationship between F_{pot} and fish weight in the Western area was not significant (P=0.188) but the Southern area was very atypical showing a significant (P<0.0001) negative slope. One possible explanation for the negative slope may be a failure to reject large spawning fish because the latter tend to spawn before the population average. The slides used to select pre-spawning fish for this area will be sent to CEFAS, Lowestoft for validation.

Annual and Intra-Annual Variation in I_{atr} and P_{rev}

To make this comparison the data for each year were regrouped to give four equal sampling periods with, as far as possible, an even allocation of samples per period (appendix Table 12). Because of this step the values of I_{atr} and P_{rev} in this review are not the same as those used in the survey working groups to calculate F_{atr} in 1992 and 1995. It was not possible to use the boot strap approach (Anon., 1996) to calculate variance because the numbers of fish in some periods were too low.

The presence of atresia is a binary response and so is modelled using logistic regression. The log of I_{atr} (in oocytes per gram) for fish with atresia is modelled by linear regression. Factors used in the model were: laboratory , year$_{1989,1992,1995}$, period $_{1-4}$ to adjust for P_{rev} varying through spawning and year. Terms representing ship, capture method, latitude and longitude and station number were not fitted because the data were not adequate. The model fitted was

$$\log (P_{rev}/(1-P_{rev})) = laboratory_i + period_j + year_k + laboratory.period_{ij} + period.year_{jk}$$

The analysis of deviance table (terms added sequentially) for the model is shown in the text Table below:

term	df	deviance	p value
laboratory	1	3.898	0.048
period	3	2.307	0.511
year	2	2.368	0.306
laboratory.period	3	10.494	0.015
period.year	5	13.252	0.021

The *year* term does not give a significant change in deviance so there is not significant evidence for a change in P_{rev} between years. The period.year interaction is significant indicating different patterns of P_{rev} within the three years. The estimated annual averages with SEs and 95% Confidence limits converted from the logistic scale are shown in text Table below:

Year	Prevalence	SE	95% CI
1989	0.357	0.054	0.260 , 0.468
1992	0.314	0.033	0.254 , 0.381
1995	0.262	0.028	0.210 , 0.321

The selected model to fit I_{atr} is: $\log (I_{atr}) = laboratory_i + period_j + year_k$ and the results of the analysis of variance analysis for $\log I_{atr}$ after fitting the model are shown in the text Table below:

	df	SS	MS	F	p-value
laboratory	1	29.39	29.39	21.43	< 0.001
period	3	18.69	6.23	4.54	0.004
year	2	26.31	13.16	9.59	< 0.001
Residual	211	289.46	1.37		

The *year* term is highly significant so there is very strong evidence that the intensity for fish with atresia is different in the three years. The fitted geometric mean values for I_{atr} are given for each laboratory in the four periods of all three years and for the overall annual means (text Table below):

		Period mean I_{atr}								Year mean I_{atr}			
		1		2		3		4					
Year	Laboratory	I_{atr}	SE	I_{atr}	SE	I_{atr}	SE	I_{atr}	SE	I_{atr}	SE	95% CI	
1989	SOAEFD	149.6	4.1	130.1	2.7	219.8	3.1	145.6	2.8	114.0	19.01	82.2	158.1
	CEFAS	77.9	2.7	67.8	2.0	114.4	2.2	75.8	2.0				
1992	SOAEFD	56.6	2.2	49.3	1.6	83.2	1.9	55.1	1.4	43.16	5.54	33.6	55.5
	CEFAS	29.5	1.4	25.3	1.2	43.3	1.4	28.7	0.9				
1995	SOAEFD	83.9	2.5	73.0	1.7	123.3	1.8	81.6	1.9	63.92	7.94	50.1	81.5
	CEFAS	43.7	1.8	38.0	1.4	64.2	1.6	42.5	1.5				

Although the means for the two laboratories are significantly different the overall trend showing highest levels in period 3 and the lowest in period 2 is the same.

Annual Variation in F_{real} in 1989,1992 and 1995 in the Western Area

The results for F_{pot}, I_{atr} and P_{rev} are combined with approximate se in the Table below with the estimated F_{real} (see appendix for the method to calculate variance and also a plot of realised fecundity in appendix Figure 2).

Year	Realised fecundity	approx. SE
1989	1217	79.7
1992	1377	37.9
1995	1303	36.5

Values of F_{pot}, F_{atr} and F_{real} to be used by the Assessment Working Group

The assessment working groups used the values of F_{pot} F_{atr} in columns 2-3 in the text Table below at the 1996 Assessment Working Group (Anon., 1997). A mean value of F_{pot} in 1992 and 1995 was used in these years because they were not significantly different and a standard adjustment of F_{atr} (10.2%) was applied to F_{pot} to calculate F_{real} for all the survey years.

	1996 Assessment Working Group			1997 Egg Survey Working Group		
Year	F_{pot}	F_{atr}	%	F_{pot}	F_{atr}	%
1977	1457	128	10.2	1526[2]	211[2]	13.8
1980	1457	128	10.2	1526[2]	211[2]	13.8
1983	1457	128	10.2	1526[2]	211[2]	13.8
1986	1457	128	10.2	1457[1]	211[2]	14.4
1989	1608	152	10.2	1608[1]	326[2]	20.3
1992	1511	154	10.2	1569[1]	138[1]	8.8
1995	1511	154	10.2	1473[1]	171[1]	11.6

[1] Survey values in 1986, 1989, 1992 and 1995 respectively
[2] This report.

The Mackerel Horse Mackerel Egg Production Workshop recommended that the values of F_{pot}, F_{atr} and F_{real} be revised in the light of the results of this review for the following reasons. A significant downward trend in F_{pot} from 1989 to 1995 has been shown and this should be incorporated into the stock assessment. Although the inter

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year variation in F_{pot} was not large, especially in the last two survey years, it must be borne in mind that the relationship between F_{pot} and fish weight also varies between years. If the F_{pot} is dependent on fish weight (Anon., 1993) then the mean size of fish in the population i.e. a population estimate of F_{pot} should be used. For the years from 1977 to 1983 the mean of F_{pot} over the period 1986 to 1995 (1526 eggs per gram) should be used.

F_{atr} has been shown to vary significantly between the three survey years (1989, 1992, 1995) irrespective of the extra variance arising from laboratory analytical procedures. In the latter two survey years (1992 and 1995) the sample numbers, (236 and 323 respectively) were much higher than in 1989 (146) which may partly explain the higher variance in the first assessment. In addition the atresia sampling (appendix Table 12) missed the first period (mid date 9 April) and was concentrated in the period of highest I_{atr} (51% of all samples were taken in the third and highest period; appendix tables 12 & 17). In conclusion the Working Group recommends that a mean F_{atr} from 1989 to 1995 be applied retrospectively from 1977 to 1986 and that the survey values of F_{atr} are used for the biomass estimates in 1989,1992 and 1995.

2.6   Maturity

2.6.1   Basis for the 1986 mackerel maturity ogive

The basis of the 1986 maturity ogive was reviewed by the working group after considering the following information presented by Witthames (WD 1997).

During the period 1977 to 1989 mackerel maturity ogives were prepared using an eight stage macroscopic maturity scale (Macer, 1976) to assess the proportions of males and females as immature (stages 1–2) or mature (stages 3–8) for each year class in the population. The maturity ogives were constructed from fish sampled each year from April to August inclusive, in Division VIa, south of 57°N and Divisions VIIe,f,g,h,j, by the Dutch commercial fleet and research vessels. The maturity at age was derived from the proportion of mature fish found amongst all the fish sampled irrespective of the catch weight or number of fish examined in each Division. The text Table below shows the historic maturity ogive for combined sexes as used by the Working group in 1985 and reviewed in 1996.

Fish age (years)	Percentage of population at maturity stages III-VIII
1	8
2	60
3 and older	>90

However, it was concluded (Anon., 1987a) that the 1984 year class was exceptional from this long-term average in that only 20% of the stock were mature at age 2 because:

1) The two year olds on the spawning ground in 1986 were about 3 cm smaller than the two year olds in 1985.
2) Expected number of mature 1984 year class as a percentage of the total number of spawning fish is 30% and the observed number of spawning females in the 1984 year class as a percentage of the total number is 11%.

Considering the first point it was recognised by the Assessment Working Group in 1987 (Anon., 1987a) that the 1984 year class was above average abundance (397 million individuals), but that it was not exceptional compared with some previous years (1978–1986 year classes ranged from 17 million to 735 million individuals Anon., 1990). The smaller mean size of 2 group fish found on the spawning grounds in 1986 was subsequently viewed as a change in distribution arising from an influx of small fish rather than a change in the mean for the whole year class. At the 1987 and 1988 Working Groups (Anon., 1987a;1988) the weights at age were revised upwards so that the weight at age of the 1984 year class, as two year olds, was now greater (300g) than either the long term mean (275g from 1969 to 1985) or the 1985 year class (250g).

With respect to the second point the proportion of spawning fish (11%) referred to the percentage of stage 6 running females in the population and excludes fish about to spawn (stages 3–5) or spent fish (stages 7–8). If these additional stages are included, which was the basis for the historic value of 30% spawning, the proportion of mature fish on the spawning grounds increases to 17.1% (Anon. , 1987b). The reduced maturity at age could

also be caused by changes in the population distribution as was the case with weight at age. For example in the years 1977–1984 the smallest least mature 2 group fish (25% mature at mean length 26.7 cm) were in Division VIIe (Anon., 1985) whilst the largest and most mature fish (100% mature at 32.9 cm) were found in Sub area VIII. The survey samples could reflect fish from any point in the distribution between these two extremes and could thus explain the observed change. This Working Group concluded that the low maturity at age observed in 1986 could be explained by points 1–2 above or also by biased sampling. If the decision to increase the weight at age 2 was sound it would be consistent to assume that the heavier fish are predominantly mature and therefore to adopt the general maturity ogive applied since 1977 for 1986.

2.6.2 Mackerel maturity ogives from the 1992 and 1995 surveys

Maturity at age information was provided at the Working Group on the Assessment of Mackerel, Horse Mackerel, Sardine and Anchovy obtained from the 1992 DEPM survey (Anon., 1997). However, this Egg Survey Working Group felt that the survey did not cover the juvenile areas particularly in the western Channel and was therefore not suitable as an unbiased estimate of maturity at age, because it overestimates the proportion of mature fish at age. Furthermore this maturity ogive was based only on macroscopic ovary observation and therefore probably also overestimates the proportion of mature fish especially the 2- and 3-year olds. Histological information on maturity was restricted to mature fish. No suitable data were available for 1995, because only mature females were collected. It was therefore impossible to provide a maturity ogive with a CV for either year.

2.6.3 Definition of immature and mature fish

Maturity at age ogives have been determined for stock assessment purposes (Anon 1985 & 1996) from April to August using macroscopic criteria defined in Macer (1976) prior to 1989 and Walsh et al. (1990) there after. In both cases maturity stages 1–2 were immature or resting respectively and stages >2 were mature and either close to spawning or just past spawning condition. This Working Group recommends changing this definition, because in the assessment the tuning takes place to the spawning stock biomass as estimated from the egg surveys. In this context the spawning stock biomass only includes fish which contribute to the annual egg production. Therefore fish, which are apparently maturing (maturity stage 3–5) but which do not produce any eggs because of mass atresia, should not be included. It is proposed to use histological criteria (Anon. 1996) to make the distinction between immature or aborting virgins and mature fish likely to spawn in the current spawning season. The definition of an immature mackerel is that the ovary should contain no post ovulatory follicles; or contain only oocytes less than 425 μm or occasionally there are totally atretic oocytes greater than 425μm. Sampling for the estimation of a maturity ogive should take place around peak spawning late May–June (Anon., 1985).

The criteria to define immature and mature horse mackerel will be developed from the 1998 surveys.

2.6.4 Problems in estimating maturity at age

Previous Working Groups (Anon., 1985; 1996) have recognised that sampling to estimate maturity at age should reflect the distribution of fish between the spawning and juvenile areas, which may differ dependent on age. Although it is not possible to provide this weighting, some progress towards producing a more representative sampling strategy could be made by fishing to provide information on the spatial distributions by age group. The use of a standardised trawl survey programme will assist in this task, because commercial sampling does not reflect the distribution of the population or allow for the collection of histological samples. The distribution of the adults is well reflected in the egg distribution (Anon., 1996). The distributions of immature components of the 1-, 2- and 3-year olds have to be made available before the 1998 surveys starts (see Section 6.5.3).

3 NORTH SEA EGG SURVEYS IN 1996

3.1 Spatial and Temporal Coverage

During the period 6 June–2 July 1996 Norway (R/V "Johan Hjort") and Denmark (R/V "Dana") carried out egg surveys in the North Sea to estimate the spawning stock biomass (SSB) of mackerel (Table 3.1). During this period the spawning area was covered three times. The last time the North Sea was covered extensively by several coverages during the spawning season was in 1990 (Iversen et al., 1991). During the period 1980–1984 the SSB was estimated based on several annual coverages and from 1984–1990 surveys were carried out every second year. In 1990 the Netherlands as well as Denmark and Norway took part in these investigations. In 1990

the surveys started in March because the investigations also covered the spawning of horse mackerel and some demersal species. Usually the mackerel spawn in the North Sea during the period from mid May to the end of July. During this period about 95 ship days were spent in 1990 while only 30 ship days were spent in 1996.

3.2   Sampling and Data Analysis

The data collecting and the handling of the samples were carried out according to Anon. (1994). The Norwegian stations were worked with a Bongo-20 and the Danish stations with a Gulf III sampler. The plankton samplers were towed at 2–3 knots for 5 minutes in 5m steps from 20m to the surface. The eggs were sorted from each of the sampled stations and their ages were estimated according to development stage and to the observed temperature at 5m. The development stages used in the calculations were eggs without a visible embryo (i.e. stage 1A+1B, Lockwood *et al.* (1981)). The average number of eggs produced per day per m^2 was calculated for each statistical rectangle of 0.5° lat. x 0.5° long. During the investigation the spawning area was covered three times and the egg production was calculated for the total investigated area for each of the three periods (Table 3.1).

3.3   Mackerel Egg Distribution

The distribution of daily egg production per m^2 is shown for each of the coverages in Figures 3.1–3.3. The sampled stations are also given in the same figures. The egg density was relatively low particularly during the first and third coverages. It seems that the main egg production was in the southwest part of the investigated area. This area was not properly covered during the first coverage (Figure 3.1). However, during the second and third coverages the sampled area was adjusted according to the results from the first survey and it seems that as a result the sampling was better during these surveys (Figures 3.2, 3.3). However, none of the surveys cover the spawning area totally. Therefore all the three surveys and particularly the first one underestimate the egg production.

3.4   Mackerel Egg Production and Spawning Stock Size Estimates

Based on the egg production estimates for each of the three periods the spawning curve was drawn (Figure 3.4). The parameters necessary for the calculation of egg production and SSB are given in Table 3.2.

By integrating the egg production curve the total egg production was estimated at $59*10^{12}$ eggs. By applying the weight fecundity relationship given by Adoff and Iversen (1983) this corresponds to a SSB of 84,000 tons which is close to that calculated in 1990, 78,000 tons (Iversen *et al.*, 1991). The SSB estimates based on egg surveys in the previous years are given in Table 3.3. The 1996 surveys were planned to take place at about the expected period of peak spawning. The surface temperature in the most productive area of the spawning area was 11–12°C. In 1996 the surface temperature appeared to be about 2°C lower than usual. This may have caused a delay in maturation and subsequent spawning. This may have delayed peak spawning as compared with previous years.

Atresia in ovaries from North Sea mackerel has never been investigated and there are no new data on fecundity since 1982. If the same weight fecundity relationship and atresia as observed in the Western area in 1995 (Anon., 1996) are applied the SSB in the North Sea is estimated at 90,000 tons which is 6,000 tons more than the estimate based on existing fecundity data (Adoff and Iversen, 1983). These estimated values of SSB were reported to ACFM in October 1996.

The estimated egg production and consequently the SSB are underestimated due to the incomplete coverage of the spawning area, especially during period one. The area at the south-western corner, which was not sampled during period one generated approximately 60% of the stage 1 eggs produced in period two and 40% in period three. Based on this a value of 50% was adopted for the unsampled area and the estimated egg production for period one was doubled. This resulted in the estimate for period one being the same as that in period two. This indicates that the surveys might have been carried out late in the peak spawning period. However, the relatively low temperature observed in the spawning area suggests that this is unlikely and that the peak of the spawning period really was covered (Figure 3.4). The adjusted egg production during the first period results in a 30% increase in SSB, i.e. 110,000 tons when applying the existing weight fecundity relationship (Iversen and Adoff, 1983). The Working Group considers this to be the most realistic estimate of the SSB in the North Sea in 1996.

3.5 Maturity

No new information was obtained about the maturity ogive of North Sea mackerel during these surveys, because fish of western and southern origin may also be present in the North Sea at the time of the surveys. Maturity data for North Sea mackerel must be obtained in May, before western and southern fish enter the area.

Table 3.1 Mackerel egg surveys in the North Sea in 1996

Coverage	1	2	3
"Dana" "Johan Hjort"	6 -15 June	15 - 20 June 17 - 23 June	23 June - 2 July
Midpoint	10 June	19 June	29 June
Egg x 10^{-12}	1.02	2.01	1.07

Table 3.2 Parameters and formulas used in the egg production and SSB estimates

Parameter	value/formula	Reference
Age of stage 1A+1B eggs	$Age = Temp^{-1.61} \times e^{7.76}$	Lockwood *et al.* 1981
Fecundity North Sea	$Fec. = 560 \times weight(g)^{1.14}$ (i.e. 1401 eggs/g female)	Iversen and Adoff 1983
Fecundity Western area 1995	1473 eggs/g female	Anon 1996
Atresia in Western area 1995	11.6%	Anon 1996
Sex ratio	1 : 1	as in previous years
Spawning period	17 May - 27 July	as in previous years, excl. 1990
Number of spawning days	72	as in previous years, excl. 1990

Table 3.3 SSB estimates from previous egg surveys in the North Sea

Year	1980	1981	1982	1983	1984	1986	1988	1990	1996
SSB 1000t	94	57	180	342	111	43	36	78	110

4 EVALUATION OF GENERALISED ADDITIVE MODELLING

4.1 Review the Results of the 1989, 1992 & 1995 Survey Analysis

4.1.1 Models adopted

Explanatory variables

The explanatory variables used were:
    date (in days from the 1st of January)
    latitude (in degrees)
    longitude (in degrees)
    distance perpendicular to the 200m contour (in metres)

distance along the 200m contour N-S (in nautical miles)
logarithm of bottom depth (in metres)

Mackerel

For mackerel a single stage GAM was adopted. A log link was used allowing multiplicative effects of the covariates. A Poisson error distribution was assumed.

Horse Mackerel

For horse mackerel a two stage GAM was adopted. The first stage modelled presence or absence of eggs. A logit link was used ass appropriate for binary data and a binomial error distribution was assumed. The second stage modelled abundance of eggs where present. A log link was used allowing multiplicative effects of the covariates. A gamma error distribution was assumed.

The models chosen were based on an empirical basis.

Smoothing was by spline with 4 degrees of freedom (df). Df were chosen based on observed data.

Variance of the estimate was calculated by bootstrap.

4.1.2   Problems encountered

The main problems encountered in the development and application of the models were as follows:

a. Partial coverage of the area by the surveys
b. Confounding of variation in space & time.
c. Choice of area to be used. This was finally based on the 1995 standard survey area (Anon 1994), and hence was larger than that used by the traditional method in 1989 and 1992.
d. Choice of start and end dates. These were standardised to 10/2 to 31/7 for the western area and 10/2 to 17/7 for the southern area. Different start dates were tested for sensitivity. The chosen dates were adopted as wider dates had no effect on the integrated volume. Narrower dates did have an effect and this has a bearing on the comparison with traditional methods in 1989 & 1992.
e. Presence of bias. GAMs are inherently biased, although this can be corrected. In this study bias was always negative. This is likely to be due to the high variance associated with high data amplitude, allowing the model to fit less tightly in these areas. A number of remedial approaches were examined.

i. **Increase in the df.** An optimal value of 12 df was chosen. This tends to reduce negative residuals in areas of high amplitude, but reduces precision in the fit generally and introduces negative bias in areas of low abundance.
ii. **Bias correction by bootstrap.** This appeared to be promising but was computationally intensive, particularly for the variance calculations and was not adopted.
iii. **Bias correction by regression.** This technique used the regression of the negative residuals against the fitted values to gave a correction factor for the fitted surface. This appeared to work well, inasmuch as there was a closer correspondence with the traditional egg production curves.

4.1.3   Results

Mackerel

1995 Western area

The model appeared to capture well; the south to north movement of spawning peak; the peak abundance and the westward shift of spawning in May. The production estimate was close to the traditional method.

1995 Southern area

The model appeared to capture; the lack of eggs on the Portuguese coast and the high density of eggs in the Cantabrian Sea in April. There were considerable problems due to sparser data than the western area and the

confounding of sampling in space and time. It was concluded that the data were inadequate for a spatio-temporal GAM.

1992

The model appeared to capture well the south to north movement of spawning peak in May/June and the westward shift of spawning in May. There was a suggestion of two peaks in spawning, and the GAM indicated that the start and end dates used for the traditional method may have been too narrow. The production estimate was less close to the traditional method, than in 1992. This may have been due to the atypical westward distribution which was poorly sampled. The GAMs are better able to extrapolate this trend and would be expected to give a higher abundance. Other possibilities for the discrepancy are the smaller area and narrower dates used in the traditional analysis.

1989

This survey posed considerable problems (for both methods) due to the bias in the German sampling in Biscay early in the season. To cope with this, the GAM was run without temporal parameters, allowing spatial data from later in the season to be applied to the German data. However this then caused a tendency to Overestimate later in the season. It was pointed out that no amount of statistics can improve badly designed survey technique.

Horse Mackerel

1995 Western area

The model appeared to capture; the later peak compared to mackerel; the more southerly distribution and the presence of two spawning peaks (end of March and start of June). Some discrepancy between approaches can be seen but this is not explained.

1995 Southern area

The model appeared to capture the initial high densities on the Portuguese coast in February/March. However, as in mackerel, the analysis was compromised by sparse data and the space/time confusion.

1992

The model appeared to capture one peak in late June. Again the dates used in the traditional method appeared to be too narrow and there was a problem with an absence of data in the south late in the survey period. The estimates were reasonably close to the traditional. Differences are possibly due to area and date effects.

1989

The model appeared to capture; the highest densities in may in the southern and central areas, the shift north and spreading east and west in June and the peak spawning in early June. There was a very good agreement between the tow approaches.

4.2   Application of the Method to the 1998 Survey

The Mackerel Assessment Working Group (Anon. 1996 Section 1.5) identified a number of areas of concern, namely:

> selection of df,
> selection of error distribution model,
> outer boundaries - spatial and temporal,
> choice of explanatory variables,
> existence of bias.

The rationale for the choices for the first two points is covered in the final report to the EU on the study contract. Sensitivity to date choice has been discussed and appears to be robust. No clear examination of sensitivity to spatial boundaries has been carried out. The explanatory variables were chosen after examination of a range of

possible parameters including temperature and vessel effects, however these were found to be unusable. The variables chosen seem sensible and apparently adequate.

The Working Group also highlighted that no formal test of the suitability of the GAMs chosen had been carried out and that no usable software and protocols have been produced.

The Working Group required (Section 1.5.3) that thorough testing be carried out using Monte Carlo simulation techniques. Tests of sensitivity of model specification were also required, particularly with reference to smoothing, choice of explanatory variables and error structures, and bias correction. The following section has been prepared in the light of these comments and on the basis of a proposed short study contract to bring the techniques to a state where it can be applied to the 1998 surveys.

4.2.1 Study proposal

This proposal is subject to a successful EU funding application. This Working Group considered that the project is vital for the application of the GAM analysis method to the egg surveys.

Proposal Summary

1. Develop models of real world egg distributions incorporating a variety of possible scenarios. Simulate sampling from these to reflect survey strategy as operated. Back check these sampling runs against real egg survey data and to integrate the GAM simulations with the simulations.
2. Evaluate model performance against simulated distributions for bias in point, variance and interval estimates. Correct the GAMs as appropriate to these evaluations. Test the robustness of the final models to a range of simulated real world scenarios.
3. Review the outcome of these studies against the traditional approach and for general use.
4. Produce usable, documented software.

Members of the Working Group were asked to comment on the proposal and to participate in this study, particularly to define and tune the potential variety of real world situations the surveys may encounter. This would be operated mainly through two workshops during the study.

The main aim of this study would be to assess the usability of the GAM technique with particular reference to the 1998 surveys.

Response from the Working Group

The initial project proposal has been considered by the Working Group and the following alterations suggested to the modellers.

1. It is felt that the appraisal should include the traditional method in the simulation studies so that the relative performance of the two techniques can be assessed, and an informed choice be made.
2. The suitability of a two-stage model for mackerel should be considered.
3. If possible the simulations should include some consideration of sampling design changes.
4. Some consideration of the sensitivity to placement of structural zeroes (area boundaries) should be included.
5. Software for general use should be implemented in S-plus for ease of use.
6. The most important real world scenarios for the simulation in order of priority should be;

- One or two peaks in egg production
- Westerly variation in the egg distribution
- South to north changes in peak abundance
- Variability in timing of peak spawning
- Different start and end dates for egg production
- Inclusion of large areas of low egg production outside the standard area

The Working Group also felt that some consideration of the use of the refined models to improving choices in effort allocation in time and space would be very useful. Particularly with reference to;

- Effect of reduced sampling intensity in time and space. E.g. in relation to modifications of survey strategy as a result of vessel breakdown etc.
- What sampling design would work best with a GAM analysis
- The effect of large gaps in survey coverage for various reasons
- Can the approach be modified to cope with some degree of spatio-temporal confusion

5   NORTH EAST ATLANTIC MACKEREL ASSESSMENT

5.1   Combining the Egg Production Estimates, Western and Southern Areas

The 1996 MHSA Working Group (Anon., 1997) recommended that methods for combining the mackerel egg survey data from the western and southern areas should be examined in order to produce a single estimate of mackerel egg production for the North East Atlantic Mackerel.

The peak of spawning of mackerel in the southern area occurs before the peak of spawning in the western area. Because there are two peaks two different estimations of the egg production curve for the two areas are necessary. Therefore, it is difficult to produce a single egg production curve for the North East Atlantic mackerel. The egg production curves constructed from the 1995 egg surveys appear to be correct and combining both estimates does not create any problems in estimating total egg production.

The GAM analysis of the 1995 egg survey in the western area indicates a peak at day 150 and a subsidiary peak at day 100. Peak egg production in the southern area is also at day 100. Therefore it is possible that two processes exist: one with a peak at day 100 (southern component) and the other with a peak at day 150 (western component). The two components may have different biological parameters (maturity ogive, fecundity and atresia). Thus, the separation between the two current components should be based on biological data, especially in the Bay of Biscay where both components are present.

5.2   Combining the Fecundity Estimates, Western and Southern Areas

The variance of the fecundity was not significantly different in the southern and western areas. The main problem in combining the fecundity for the two areas could be the different relationship between the eggs per gram and the fish weight. Fish weight has a significant effect on fecundity. The area effect is not significant once the weight effect has been considered. The western mackerel data show a trend of fecundity (in eggs per gram) increasing with weight but the trend is not significant. On the other hand, the southern mackerel data show a significant decreasing trend of fecundity with weight. It is necessary to check the southern area data (Witthames and Maxwell, WD1997).

These differences are likely to be bigger between individuals spawning in the Cantabrian Sea (Southern component) and those spawning in the Celtic Sea (Western component). Differences may also occur in the Bay of Biscay between individuals spawning either in the Armorican slope or on the Cantabrian coast.

5.3   Combining the Atresia Estimates, Western and Southern Areas

No atresia observations have been made in the southern area, so it is not possible to combine estimates.

5.4   Combining Estimates of the Maturity Ogives

For combining the maturity ogives the biomass of the spawning fraction of the western and southern areas should be used as weighting factor.

6   PLANNING OF THE 1998 MACKEREL AND HORSE MACKEREL EGG SURVEYS IN THE WESTERN AND SOUTHERN AREAS

6.1   Countries and Ships Participating

England, Germany, Ireland, Netherlands, Scotland, Portugal, Spain, Spain/Basque Country and Norway will participate in the mackerel/horse mackerel egg surveys in the western and southern area in 1998. Survey

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coverage of the western and southern areas (Figure 6.1) will be even more closely interlinked than in 1995 (Table 6.1).

The survey will be split into seven sampling periods, allowing full coverage of the expected spawning area (periods 1–6) and 5 of the western area (periods 3–7) (see Table 6.1). The widest area cover is provided during the third sampling period when the distribution of mackerel and horse mackerel spawning is at its most widespread in the southern area. For this period an overlap of the sampling areas is planned for the Spanish and German surveys, in order to ensure a complete coverage of the southern area at the time of peak spawning. For this purpose a flexible spatial coverage, into the southern area and at the north-western edge of the survey area, is allocated to the German survey. The details of the coverage of the Cantabrian Sea will be coordinated by direct communication between the RV "Cornide de Saavedra" and RV "Walther Herwig III" when operating in the area.

In the western area maximum deployment of effort is during the fourth, fifth and sixth sampling periods, the latter two coincide with expected peak spawning of mackerel and horse mackerel in the area. In order to achieve maximum coverage of the western area in each sampling period the Scottish survey in the fourth sampling period will attempt to cover the entire area from north to south omitting every second transect. In the fifth sampling period the transects previously omitted will be sampled from the south to north. The same sampling strategy will apply to the Norwegian survey in the fifth and sixth sampling periods.

Two vessels will be operating in the Cantabrian Sea and the southern part of the Bay of Biscay in the fifth sampling period. Again the details of coverage will be coordinated between the RV "Tridens" and the Basque charter vessel. It is hoped that the seventh and final sampling period in the western area is covered by a Scottish charter vessel in the first three weeks of July. The core sampling area is set between latitudes 46°N and 53°N.

Deployment of research vessel effort is shown in Table 6.1 while proposed area coverages by nation during each survey period are shown in Figures 6.2–6.8 and Table 6.2.

Countries should report changes to the ship's deployment schedule as soon as possible to John Nichols (CEFAS, Lowestoft). This will allow any resultant problems to be addressed in good time and potential solutions explored.

6.2   Sampling Areas and Sampling Effort

As in previous years it was decided that the spatial and temporal distribution of sampling would be designed to ensure an adequate coverage of both mackerel and horse mackerel spawning and that estimates of stage 1 egg production would be made for both species.

Since the surveys were started in 1977 considerable changes have been made to the standard sampling area and these have been described in Section 8.4 (Anon. 1994). In 1995 changes were made to the western boundaries because of the unusual westerly distribution of mackerel eggs which occurred in period 3, 1992. The distribution of the stage 1 eggs was therefore examined again for the 1995 surveys (Anon., 1996) to determine whether the additional rectangles covered the main spawning areas of mackerel and horse mackerel. A summary of the coverage is a follows:

Mackerel

The coverage during period 1-3 appeared to be adequate and no additional sampling stations are necessary.

In periods 4 and 5 coverage was inadequate along the western boundary and extra stations are required between 46° and 48° 30' and between 50°30' and 51°. It is also possible that the area between 45° and 46° 30' that lies west of the western boundary may contain important concentrations of eggs during this period Vessels covering this area should therefore ensure that stations along Row 20 do not contain eggs before leaving the area. (see Section on sampling strategy).

Coverage appears to have been adequate during periods 6 and 7 - although there is some indication that small concentrations of eggs are found north of 58°. This area should be examined by vessels if the opportunity arises as vessels enter or leave the survey area.

<u>Horse mackerel</u>

In general the spawning concentrations of horse mackerel seem to have been reasonably well contained within the standard area.

In period 4, 5 and 6 additional sampling is required between 45° and 48° 30'N for the same stations as those considered necessary for mackerel.

The new standard area is shown in Figure 6.1 with the proposed additional rectangles on the western edge shaded.

6.3   Recommendations of the Plankton Sampler Study Group

A progress report was given on the studies carried out under an EU Concerted Action Contract to investigate high speed plankton sampler design, flow measurement and calibration. No recommendations have yet been provided by the Plankton Sampler Study Group to be incorporated into the field programme for the 1998 surveys.

Considerable progress has been made, by the use of a Laser / Doppler system in a flume tank, in measuring the sampling efficiency of the Gulf III and 20 cm Bongo designs. This work has resolved the large differences in the interpretation of earlier calibrations which used different primary calibration devices. The experimental work has now been supported by modelling studies which have examined the effects on efficiency of small changes in the configuration of the Gulf III nose cone.

Two non intrusive methods of measuring flow into plankton samplers have been tested in a flume tank as a part of the Concerted Action studies. Sea trials of one of these, a "time of flight acoustic device", will be carried out in March 1997. The development and calibration of this device will not be completed in time to make it commercially available for use in the 1998 surveys. Further development on the other device, an electromagnetic flowmeter, has been suspended.

The results of all these studies are currently being described in the Final Report to the EU. A summary of the conclusions and recommendations will be presented at the 1997 ICES Annual Science Meeting. This will include advice on the inherent efficiency of the various national designs of Gulf III sampler and the Bongo sampler. It will also detail the way in which the flowmeter readings should be used to calculate volume of water filtered by these samplers. This advice should be used for the calculation of volume filtered during the 1998 surveys.

6.4   Sampling Strategy, Gear and Procedures

A manual for the conduct of egg surveys, targeted at the AEPM, is given in Section 8 of the Report of the Mackerel/Horse Mackerel Egg Production Workshop (Anon., 1994). Those instructions are repeated in Sections 6.4.1 to 6.4.8. Any changes, additions or clarifications, to the instructions in the 1994 manual, have been underlined in this report.

6.4.1   Sampling gear

The standard samplers acceptable for use on the 1998 surveys are national variants of the Gulf III or towed Bongo samplers.

The Gulf III sampler is deployed on a double oblique tow, at 5 knots, from the surface to sampling depth and return, and the Bongo sampler at 2–3 knots. The aim is for an even, not stepped, dive profile filtering the same volume of water from each depth band.

Although a mesh size of 500 micron aperture is adequate for sampling mackerel and horse mackerel eggs, a nylon mesh with an aperture between 250 and 280 microns is the recommended size for these surveys. This allows the plankton samples to be more widely used for investigations on other species and taxa. If serious clogging occurs then a change to a 500 micron aperture mesh can be made (this change has only rarely been made on any of the surveys).

<u>The aperture on the Gulf III type samplers should be 20 cm diameter in order to ensure that an adequate volume of water is filtered to quantitatively sample the eggs of other species, in particular hake, which may be present at lower densities than the target species.</u>

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The aperture for the Bongo samplers should be either 40 cm or 60 cm diameter.

6.4.2 Target species

The sampling programme for 1998 will be targeted at mackerel and horse mackerel. An egg production estimate will be calculated for both species in both areas. In addition an egg production estimate for mackerel will be calculated for the combined North East Atlantic area.

6.4.3 Standard sampling area

Changes to the standard sampling area for the 1998 surveys are described and defined in section 6.2 of this report. Additional rectangles have been added to the standard area as a result of the changes in the distribution of mackerel and horse mackerel eggs noted in the 1995 survey. A total of eight rectangles have been added at the northern end of the survey area and a further twenty four rectangles on the western edge between latitudes 45°30'N and 51°N (Figure 6.1)

6.4.4 Sampling strategy

The sampling strategy in the western and southern areas in 1998 will be targeted at the AEPM only. From analyses of 1992 egg survey data presented to the 1994 Egg Production Workshop (Anon., 1994) and from knowledge of previous years distributions it is clear that egg distributions in all survey periods conform to a characteristic spatial pattern which can be modelled. These analyses indicated that changes in the distribution of sampling effort coupled with the use of a model based approach could lead to significant improvements in estimates of egg production in future. From the point of view of sampling effort the analysis indicated that two important factors needed to be considered when planning survey strategy.

Firstly, a set of rules must be established in order to decide when to stop sampling along a given transect in order to ensure that the whole area of egg distribution is sampled with no effort is wasted outside the spawning area.

Secondly, some guide-lines need to be provided to cruise leaders on the number and spacing of transects which may be omitted in order to best match available effort to the size of the area to be surveyed. This approach was adopted for the 1995 surveys and it is proposed that the same flexible approach be adopted for the 1998 surveys. This will permit an alternative analysis of the data set using a GAM as discussed in Section 4.

As a first guide to planning the distribution of sampling effort in the western area and southern areas in 1998, historic egg distribution data are provided in Figures 6.2–6.8 The core distributional areas, identified for each of the different sampling periods, should always be sampled to the north/south and east/west limits although individual transects may be omitted. When sampling along transects, shipboard enumeration of results should be undertaken several rectangles before the limit of the core area is reached. Sampling should be completed either after one zero or (near zero) value or two consecutive low values i.e. less than about 20 stage I eggs of either species present in the sample. In practice eggs do not become visible until an hour or so after fixation - roughly the steaming time between stations - so that one extra station after a zero or 2 low values will always be necessary before steaming to the next transect. In some cases it will be necessary to sample beyond the core area limits and even beyond the standard survey area limits.

With regard to the spacing and omission of sampling transects this will depend on the size of the area to be covered and the amount of ship time available. During periods when several ships are available it should be possible to sample all transects while at other times it may be necessary to omit several, at least during the first pass over the designated sampling area. No more than three consecutive transects should ever be omitted. Given that the area to be covered is more or less known, as is ship time, cruise leaders should be able to estimate fairly accurately the number of full transects they will be able to make. **It is strongly recommended that, where practicable, and even where total coverage is expected, a first pass over the area be made on alternate transects. The intervening transects should be sampled on the return leg.** In this way weather problems, equipment failures and vessel breakdown need not seriously prejudice results. Such a strategy, furthermore, enables a better evaluation of distributional change with time which is likely to be important in modelling the results. An example of an appropriate sampling strategy where only one in three transects can be fully sampled is given in Figure 6.16 in Anon. (1994).

A flexible approach will again be adopted to replicate sampling within a rectangle. Additional sampling should be carried out in areas where high densities of either mackerel or horse mackerel eggs can be expected. As guidance to the areas where these high densities are likely to occur, cruise leaders should refer to the charts showing the maximum contribution to egg production of either species in each time period (Figures 6.2–6.8). In order to improve spatial resolution, replicate samples within a rectangle should not be taken in the centre of those rectangles but should be evenly spaced in an east-west direction.

6.4.5 Sampling depth

Maximum sampling depth is to **200 m** or to within **2 m of the bottom** where the bottom depth is less than 200 m. In the presence of a thermocline greater than **2.5°C in 10 m depth**, sampling can be confined to a maximum depth of **20 m below the base of the thermocline**.

For subsequent sample analysis the conversion, from numbers per m^3 to numbers beneath a m^2, uses the **maximum sampled depth**. This protocol has operated throughout all the surveys.

6.4.6 Sample fixation

The standard fixative for use on these surveys is a 4% solution of buffered formaldehyde in either distilled or freshwater. This solution is approximately isosmotic with sea water and should be used in preference to a 4% formaldehyde solution in sea water in order to minimise the problem of damage and distortion. The sample should be directly fixed with the addition of the 4% formaldehyde solution and should not come into contact with formaldehyde strength in excess of 4%.

The 4% solution should be made up as follows; 40% formaldehyde as purchased, 1 part; distilled or freshwater, 9 parts; plus an appropriate buffer to pH 7–8.

The volume of plankton in a sample jar must never exceed 50% of the volume of the jar. Excess sample should be fixed separately in additional jars. Details of an alternative fixative, giving better definition of egg development stage, for a more precise estimate of elapsed time since spawning, were given in Anon. (1988). That fixative is ethanol (95%), 9.5 parts; formalin (10%), 1 part; glacial acetic acid, 0.5 parts.

6.4.7 Sample sorting, egg identification, staging and ageing

Whenever practicable the whole sample should be sorted in order to remove all the eggs of non target species such as hake and sardine, which may be present in lower densities than the target species. All sorted eggs should be kept in tubes, in fixative, inside the sample container for future reference and use. Only the eggs of mackerel and horse mackerel need be identified to species. A minimum of 100 eggs of each of the target species must be staged from the sorted sample or sub-sample.

The eggs of mackerel should be classified into one of five morphological stages (I, II, III, IV and V) (Lockwood et al., 1981) following the development criteria described for plaice (Simpson, 1959). For horse mackerel the description of stages is the same with the exception of stage V which does not exist. Horse mackerel larvae hatch at the end of egg stage IV (Pipe and Walker, 1987).

For the estimation of daily egg production for both species only the counts of stage I eggs are used. This is recognised as a conservative estimate of the total spawned because some mortality probably occurs during development. However until there is consistency, between all countries, in the identification of the other stages (see Section 2.1) the other stages cannot be used for the estimation of total eggs spawned.

To convert abundance of eggs into daily egg production, data on the rate of development is required. For mackerel the relationship between egg development rate and temperature was described by Lockwood et al., (1977, 1981). This has been used as the basis for calculating daily production of stage I eggs on all the surveys from 1977. For horse mackerel similar egg development data are given by Pipe and Walker (1987) and have also been used for the calculation of stage I egg production since 1977.

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The formula for calculating the age of **stage I mackerel eggs** from the sea temperature (T°C) is:

$$\text{Log}_e \text{ time (hours)} = -1.61 \log_e (T°C) + 7.76$$

For calculating the age of **stage I horse mackerel** eggs the formula is:

$$\text{Log}_e \text{ time (hours)} = -1.608 \log_e (T°C) + 7.713.$$

When available the temperature at 20 m depth should be used for the calculation of egg stage duration. If that is not available then the sub-surface temperature (ca. 3 m) should be used.

6.4.8   Rectangle sampling

The protocol is as follows. In order to qualify for an interpolated value an unsampled rectangle must have a minimum of two sampled rectangles immediately adjacent to it. Once qualified the sampled values of all surrounding rectangles, both immediately adjacent and diagonally adjacent are used to calculate the interpolated value. The interpolated value is the arithmetic mean of all those surrounding rectangles.

Once calculated, interpolated values are not used in order to calculate values for other unsampled rectangles, or to qualify those rectangles for interpolation. No values are to be extrapolated outside the sampled area.

On some occasions, and in particular where multiple observations are made within a rectangle, for example the CalVET net sampling by Spain, sampling positions may fall on a dividing line between rectangles. When this occurs the sample is allocated to the rectangle to the north of the line of latitude and to the west of the line of longitude.

6.5   Biological Sampling for Fecundity, Atresia and Maturity

6.5.1   Sampling for total fecundity

Mackerel

Western area

England will collect samples for total fecundity studies between 6 March and 6 April 1998 during the CEFAS western Channel Groundfish Survey. Sample jars filled to a standard weight with either 0.1M phosphate buffered pH7.0 4% formaldehyde or Gilson fixative (Simpson, 1951) will be prepared at Lowestoft for the Cirolana collection. A total of 150 fish should be collected covering the length range of 27 cm and above. This will correspond to about 10 fish per cm. Only fish in late pre-spawning stage 3 should be collected (Walsh *et al.*, 1990). Ovaries should be carefully dissected out of the fish. The ovary membrane should be pierced to allow penetration of the fixative to the lumen. One ovary should be placed in buffered formaldehyde and the other in Gilson's fluid (minimum 2x ovary volume). At the laboratory the ovary fixed in formaldehyde should be prepared for wax histology, sectioned and stained (H&E) to exclude spawning fish from the sample. Length and weight of each fish and gonad weight should be recorded. Results should be presented at the next Egg Survey Working Group as in Table 5.8.1 from Anon. (1996) except for the atresia data.

Southern area

Spain will collect a total of 150 fish in February 1998, covering the length range of 22 cm and above. This will correspond to about 10 fish per cm. Only fish in late pre-spawning stage 3 should be collected (Walsh et al., 1990). Ovaries should be carefully dissected out of the fish. The ovary membrane should be pierced to allow penetration of the fixative to the lumen. One ovary should be placed in buffered formaldehyde and the other in Gilson's fluid (minimum 2x ovary volume). The ovary fixed in 0.1M phosphate buffered pH7.0 4% formaldehyde (minimum 2x ovary volume) should be prepared for wax histology, sectioned and stained (H&E) to exclude spawning fish from the sample. Length and weight of each fish and gonad weight should be recorded and the otoliths taken. The fecundity study will be carried out at IEO, Vigo. Results should be presented at the next Egg Survey Working Group as in Table 5.8.1 from Anon. (1996) except for the atresia data.

Horse Mackerel

Western area

Netherlands should collect samples for total fecundity estimation as early as possible in April. A total of 150 fish should be collected covering the length range of 22 cm and above. This will correspond to about 10 fish per cm.

Only fish in late pre-spawning stage 3 should be collected (Walsh et al., 1990). Ovaries should be carefully dissected out of the fish. Both ovaries should be placed in 0.1M phosphate buffered pH7.0 4% formaldehyde (minimum 2x ovary volume). Length and weight of each fish and gonad weight should be recorded. These ovaries should be sent to RIVO-DLO in IJmuiden, the Netherlands, where they will be used for the preparation of resin slides for histological analysis. Results should be presented at the next Egg Survey Working Group as in Table 5.8.1 in Anon. (1996).

Southern area

Portugal should collect samples for total fecundity studies in January and Spain in March. Each country should collect 75 fish to ensure that a total of 150 fish are collected covering the length range of 22 cm and above. This will correspond to about 5 fish per cm per country. Only fish in late pre-spawning stage 3 should be collected (Walsh et al., 1990). Ovaries should be carefully dissected out of the fish. Both ovaries should be placed in 0.1M phosphate buffered pH7.0 4% formaldehyde (minimum 2x ovary volume). Length and weight of each fish and gonad weight should be recorded and the otoliths taken. These ovaries will be used for the preparation of resin slides for histological analysis. The fecundity study will be carried out by both IPIMAR, Lisbon and IEO, Vigo. Results should be presented at the next Egg Survey Working Group as in Table 5.8.1 in Anon. (1996).

6.5.2   Sampling for atresia

Mackerel and Horse Mackerel

Western and Southern area

For the estimation of prevalence and relative intensity of atresia both mackerel and horse mackerel ovaries from a minimum of ninety mature fish should be collected from each survey period and survey area as given in Table 6.1

It is recommended that a midwater trawl, fished close to the surface in the dark or fished close to the bottom during day light, is used to sample the population. The first fifteen randomly selected mature (maturity stages 3–6 (Walsh et al., 1990)) females should be taken from six locations, spaced along the north-south axis of the egg survey area, close to the 200 metre contour along the shelf edge. Ovaries should be dissected out without damage to the outer wall of the ovary and fixed in a minimum of two volumes of 4% formaldehyde, 0.1M phosphate buffered to pH 7, for subsequent histological analysis. These will be used for the preparation of resin slides for histological analysis. A selection of a maximum of fifty fish per period for atresia analysis is necessary. Only fish in spawning condition (histological markers include presence of migratory nuclei, hydrated oocytes and post ovulatory follicles) should be included in this selection.

The numbers of fish to be collected by area and period are given in Table 6.3 which also shows to which Laboratory they have to be sent. The sampling co-ordinators are listed in the table.

The atresia results should be presented in a similar format as given in Tables 5.8.2 and 5.8.3 of Anon., 1996 to the next meeting of the Mackerel/Horse Mackerel Egg Survey Working Group.

It is recommended that ten slides are circulated at the beginning of 1998 to check the interpretation and estimation criteria before the general work commences (see Section 2.3).

6.5.3   Sampling for maturity at age

In the light of the problems in estimating the maturity at age (see Section 2.6.4) the following sampling scheme has been proposed by the Working Group for the 1998 egg survey for both the western and southern areas. Trawling for both mackerel and horse mackerel should be carried out, both in the adult and juvenile areas, to estimate the maturity at age from histological slides of ovaries. The time available to carry out trawl hauls during the egg survey will be limited, because the first priority will be plankton sampling. It was agreed that samples for maturity estimation would only be taken around the peak of spawning when the largest part of the spawning population is expected to be present (Anon., 1985 and 1993). It is assumed that by that time, very few of the old fish will have left the spawning area, and that most of the young fish will have migrated into it. During each coverage the ovary samples should be proportionally collected over both the adult area, according to the egg production, and in the juvenile area, according to the distribution of 1, 2 and 3 year old fish.

This Working Group recommends that information on the distribution of 1, 2 and 3 year old mackerel and horse mackerel be made available to the Working Group on the Assessment of Mackerel, Horse Mackerel, Sardine and Anchovy for its next meeting in September 1997. The Working Group is asked to provide charts indicating the proposed distribution of trawl hauls over the egg survey area and the juvenile areas.

Mackerel

Around peak spawning time a total of twenty trawl hauls should be made, supplemented by additional hauls as necessary, to sample maturity. These trawl hauls should be distributed over both the western and southern area in such a way that they are representative of the distribution of the adult and juvenile mackerel.

Horse Mackerel

Around peak spawning time a minimum of fourteen trawl hauls in the southern area and fourteen hauls in the western area should be made. In each of the areas the trawl hauls should be distributed in such a way that they are representative of the distribution of the adult and juvenile mackerel.

Mackerel and Horse Mackerel

From each trawl location a total of one hundred immature and mature females should be taken at random from the catch. Length, weight and visual maturity stage of each fish should be recorded and the otoliths taken, because the proportion mature has to be estimated by age group. Fish which have ovaries with many clear hyaline eggs or which have eggs in the lumen should clearly be indicated on the input form (Table 6.4)

Ovaries should be dissected out whole from the fish and placed in a minimum 2x ovary volume of 0.1M phosphate buffered pH 7.0, 4% formaldehyde. Different size jars, 25, 50, 200 ml., are recommended for small medium and large ovaries to save space and freight costs. The fixed ovary samples have to be sent to the laboratories involved in the analysis of each area. They are listed, together with the names of the survey co-ordinators, in Table 6.5. These ovaries will be used either for the preparation of wax slides for histological analysis or, if maturity is not in doubt (presence of hyaline eggs or ovaries approximately < 2 g), they will be held for later reference.

Results should be presented to the next Mackerel/Horse Mackerel Egg Survey Working Group meeting as in Table 6.4.

6.6   Data Analysis

To convert the number of eggs counted in each sample or sub-sample to the number of eggs per m^2, the following calculations are made. Firstly the volume of sea water filtered by the sampler during the haul is calculated.

Volume filtered (m^3) =

$$= \frac{\text{Flowm-revs x Aperture}}{\text{Flowm-calibr.}} \times \text{Efficiency Factor}$$

The number of eggs/m^2 is calculated from the formula:

$$\text{Eggs/m}^2 = \frac{\text{Eggs counted x Factor}}{\text{Volume Filtered (m}^3)} \times \text{Depth Sampled}$$

Where:

Flowm-revs.	=	Number of revolutions of the flow meter during a tow.
Aperture	=	The area of the mouth opening of the sampler in m^2.
Flowm-calib.	=	The number of flow meter revolutions per metre towed, obtained from the flume or sea calibration in free flow.
Eggs counted	=	Number of eggs in the sub-sample.
Factor	=	Raising factor from the sub-sample to the whole sample.

Depth sampled   =   The maximum depth of the sampler during the tow in metres.

Efficiency Factor =   The sampler efficiency from flume or towing tank calibration.

Numbers of eggs per m^2 are raised to numbers per m^2 per day using development equation for both species (see Section 6.4.7) in the following way:

For stage I **mackerel** eggs:

$$Eggs/m^2/day = \frac{24 \times Eggs/m2}{exp[-1.61 \log_e(T^\circ C) + 7.76]}$$

For stage I **horse mackerel** eggs:

$$Eggs/m^2/day = \frac{24 \times Eggs/m2}{exp[-1.608 \log_e(T^\circ C) + 7.713]}$$

$Eggs/m^2/day$ are then raised to the area of the rectangle they represent. The rectangle values are summed to give numbers of eggs per day in each stage over the survey area for each sampling period. Rectangle areas are calculated by each $^1/_2^\circ$ row of latitude using the formula:

$$Area (m^2) = (\cos(latitude) \times 30 \times 1853.2) \times (30 \times 1853.2)$$

The next stages in the estimation of annual egg production are:

- estimating the daily egg production for each survey period in turn
- integrating the daily egg production histogram, to give annual egg production
- calculating the variance of the estimate of annual egg production

The method was modified for use in the analysis of the 1995 survey data. It is fully described in Section 5.3.3 of the report of those surveys (Anon., 1996b). The same methods used for these analyses will be used for the analysis of the 1998 survey data.

6.7   Co-ordination, Communication, Deadlines and Reporting

The co-ordinator of the 1998 western egg survey will be J.H. Nichols, CEFAS, Lowestoft Laboratory, Pakefield Road, Lowestoft, Suffolk, UK. NR33 0HT.

The co-ordinator of the 1998 southern egg survey will be F. Borges from IPIMAR, Lisbon, Portugal.

Participants, who will be surveying during the same time period, should contact each other prior to their cruises to co-ordinate strategies and areas of overlap if any. Co-ordinators will obtain and provide details of vessels communication systems for use in maintaining regular contact during surveys. Contact with cruise leaders from the previous survey is also recommended to give prior indication of any distributional abnormalities.

Data input forms for the survey results and charts showing the proposed trawling positions for sampling maturity will be despatched to all participants by the area co-ordinators after the meeting of the MHSA Working Group in September 1997.

The co-ordinator of the western egg survey data base will be Julie Mc Millan, Marine Laboratory, PO Box 101, Victoria Road, Aberdeen AB9 8DB, UK.

The co-ordinator of the southern egg survey data base will be A. Solá from the Instituto Español de Oceanografía, Madrid, Spain.

J. Mc Millan and A. Solá will be responsible for loading data onto the data base, checking their validity and estimating stage I densities. The data base will be available to all participants in the survey.

30 September 1998 is the deadline for sending the egg survey results of both mackerel and horse mackerel to J. Mc Millan and A. Solá.

The deadline for the analysis of all the samples and data relating to the adult parameters, collected during the 1998 surveys, is 15 March 1999.

The next meeting of the ICES Working Group on Mackerel and Horse mackerel Egg surveys is proposed to be held from the 13–19 April 1999 in Hamburg, Germany. It is proposed that the stock assessment biologists for the mackerel and horse mackerel stocks attend the last two days of this meeting. This will allow the finalised estimates of SSB, from the 1998 egg surveys to be used to re-tune the VPA estimates of stock size in time for consideration by ACFM at their May meeting. It is important that the meeting does not coincide with meetings of either the Herring or Mackerel, Horse Mackerel, Sardine and Anchovy Working Groups.

7   PLANNING OF FUTURE NORTH SEA EGG SURVEYS

The ship time put into the egg surveys in the North Sea was significantly reduced in 1996 compared to the previous survey in 1990. This is probably because no signs of improvement of the stock have been observed since 1990. However, in the autumn/winter of 1996/1997 small mackerel (1996 year class) have been observed in the North Sea for the first time in many years (Iversen, pers.com.). This means that there might be hope for an improvement in the spawning stock the coming years. There will be more information available about the 1996 year class at the Assessment Working Group in September 1997.

This Working Group recommends that a new egg survey should be carried out in the North Sea in 1999, when the 1996 year class is expected to be fully recruited to the SSB.

8   DEFICIENCIES AND RECOMMENDATIONS

1. The Working Group recommends that a new egg survey should be carried out in the North Sea in 1999, when the 1996 year class is expected to be fully recruited to the SSB;

2. The Working Group recommends that a further egg exchange exercise, designed to improve the precision of identification and staging of both mackerel and horse mackerel eggs, should be organised for the 1998 surveys by S.P. Milligan (UK, England and Wales);

3. The Working Group recommends that information on the distribution of 1, 2 and 3 year old mackerel and horse mackerel should be made available to the Working Group on the Assessment of Mackerel, Horse Mackerel, Sardine and Anchovy at the meeting in September 1997. This Group will then provide charts which will show where trawl hauls should be distributed over the egg survey area and the juvenile areas;

4. The Working Group recommends that an exchange of histological atresia slides should take place between institutes (CEFAS, Aberdeen University, IEO, IPIMAR for mackerel and RIVO-DLO, IEO, IPIMAR and FRC for horse mackerel) The exchange should be coordinated by Mr Witthames (CEFAS) for mackerel and Mr Vingerhoed (RIVO) for horse mackerel;

5. The Working Group recommends that all fish eggs and, if possible fish larvae, should be extracted from the samples collected in the 1998 surveys and made available to the relevant institutes;

6. The Working Group recommends that full support should be given to the proposed EU funded project on validation of Generalised Additive Models (GAMs) for the analysis of the mackerel and horse mackerel surveys;

7. The Working Group recommends that the next meeting of the Group should take place in Hamburg from 13 April–19 April 1999. It is important that this meeting should be attended for the last two days by the relevant stock assessment biologists. It is also important that the meeting does not coincide with meetings of either the Herring or Mackerel, Horse Mackerel, Sardine and Anchovy Assessment Working Groups.

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Table 6.1   Proposed deployment of research vessel effort in th Western and Southern survey areas in 1998.

Sampling Period	Country	Area	Ship	#	Period	Survey mid-pont	Latitude to be covered
1	Portugal	South	Capricornio	2	12-25 Jan	19-Jan	36°N-43°N
2	Portugal	South	Capricornio	2	09-22 Feb	15-Feb	36°N-43°N
3	Portugal	South	Capricornio	3	09-21 March	15 March	36°N-43°N
3	Germany	West/South	Walther Herwig III	2	07-29 March	18 March	44°N-52°N
3	Spain	South/West	Cornide	2	23 March-06 Apr.	29 March	42°N-47°N
4	Spain	South/West	Cornide	2	20 April-04 May	27 May	42°N-47°N
4	Netherlands	West	Tridens	3	14 April-01 May	22 May	47°N-49°N
4/5	Scotland	West	Scotia	3	27 Apr.-18 May	07 May	49°N-58°N
5	Spain/Basque	South/West	AZTI charter v.	2	10-25 May	17 May	44°N-47°N
5	Netherlands	West/South	Tridens	3	11-29 May	20 May	43°N-49°N
5/6	Norway	West	G.O.Sars	3	25 May-15 June	04 June	49°N-58°N
6	Ireland	West	Celtic Voyager	3	1-22 June	11 June	49°N-58°N
6	England/Wales	West	Cirolana	3	5-29 June	16 June	43°N-49°N
7	Scotland	West	Charter Vessel	3	ca.29 June-20 July	ca. 10 July	appr. 44°N-54°N

Table 6.2 Survey schedule

# Stat./Lat.			Mackerel / Horse Makerel Egg Survey Planning Group, Lisboa 1997 — Coverage of area (Latitude - Latitude)																							Sampling Period
			20	13	8	8	10	10	8	27	36	18	31	43	37	31	31	28	24	24	21	13	10	10	5	
Month	Week	Week	36	37	38	39	40	41	42	43	44	45	46	47	48	49	50	51	52	53	54	55	56	57	58	

Body annotations (as they appear across the grid):

- **Jan.** Weeks 3–5: *Portugal 12-25 January — 13 days/77 Stations=6 st/d* — Sampling Period **1**
- **Feb.** Weeks 6–10: *Portugal 9-21 Febuary — 12 days/77 Stations=6 st/d* — Sampling Period **2**
- **March** Weeks 11–13:
  - *Portugal 09-22 March — 13 days for 77 st. = 6 st./d*
  - *Germany 07-29 March — 20 fishing days for approx.255 st = 13 st/d*
  - Sampling Period **3**
- Weeks 13–14: *Spain 23 March until 06 April* ; *Esp: 14 d. 120 st.= 9 st/d* ; *Scotl. 1st half: apr.80st/10 days=8 st/d (every 2nd transect)*
- **April** Weeks 14–18:
  - *Scotl.: 2nd half: appr.80 st./11 days= 7 st/d*
  - *Netherl. 14 April-01 May — 12 fishing days for 111 st = 9 st/d*
  - *here every second transect alternating with Scotl. and Norway*
  - *Spain 20 April until 04 May* ; *Esp: 14 d. 120 st.= 9 st/d*
  - *Scotland 27 April - 06 May*
  - Sampling Period **4**
- **May** Weeks 19–23:
  - *AZTI: 14 days for 64 st. = 5 st./d*
  - *NL: 14 fishing d.for appr.128 st. = 9 st./d*
  - *peak period of mackerel, horse mackerel spawning in western area*
  - *AZTI 10-25 May (46°30´) 1-5°W*
  - *Netherlands 11-29 May*
  - *Scotland 07 - 18 May* ; *N: 20 days for 197 st.=10st/d*
  - *Norway 25 May - 15 June*
  - Sampling Period **5**
- **June** Weeks 24–26:
  - *England & Wales 5-29 June — 22 fishing days for 223 st. = 10 st/d*
  - *Ireland 1-22 June — IRL: 21 fishing days for 166 st.= 8 st/d*
  - *here every second transect alternating between Noway and Irland*
  - Sampling Period **6**
- **July** Weeks 27–31:
  - *Scotland /Charter 29 June - 20 July*
  - Sampling Period **7**

Table 6.3 Minimum number of ovaries to be sampled for atresia investigations on each survey in 1998. During each coverage a minimum of 90 ovaries should be sampled in the southern area (S) and 90 in the western area (W) for both mackerel and horse mackerel. The countries responsible for sampling and the laboratories where the samples should be sent for histological analysis are shown in the table. The coordinators for sampling and analysis in each country are listed below the table.

COVERAGE	COUNTRY	PERIOD	AREA	MACKEREL		HORSE MACKEREL	
				MIN. NO. FISH	LABORATORY	MINIMUM NO. FISH	LABORATORY
1	Portugal	12/1 to 25/1	36N - 43N	90	IEO Vigo	90	IPIMAR
2	Portugal	9/2 to 22/2	36N - 43N	90	IEO Vigo	90	IPIMAR
3	Portugal	9/3 to 22/3	36N - 43N	45	IEO Vigo	45	IPIMAR
	Spain	23/3 to 26/4	42N - 47N	45(S) and 45(W)	IEO+CEFAS+SOAEFD	45(S) and 45(W)	IEO+RIVO
	Germany	7/3 to 29/3	44N - 52N	45	CEFAS+SOAEFD	45	RIVO
4	Spain	20/4 to 4/5	42N - 47N	90(S) and 30(W)	IEO+CEFAS+SOAEFD	90(S) and 30(W)	IEO+RIVO
	Netherlands	14/4 to 1/5	47N - 49N	30	CEFAS+SOAEFD	30	RIVO
	Scotland	27/4 to 7/5	49N - 58N	30	CEFAS+SOAEFD	30	RIVO
5	Scotland	7/5 to 18/5	49N - 58N	25	CEFAS+SOAEFD	25	RIVO
	Netherlands	11/5 to 29/5	43N - 49N	90(S) and 25(W)	IEO+CEFAS+SOAEFD	90(S) and 25(W)	IEO+RIVO
	Spain (Basque)	10/5 to 25/5	44N - 47N	25	CEFAS+SOAEFD	25	RIVO
	Norway	25/5 to 4/6	49N - 58N	25	CEFAS+SOAEFD	25	RIVO
6	Norway	4/6 to 15/6	49N - 58N	30	CEFAS+SOAEFD	30	RIVO
	Ireland	1/6 to 22/6	49N - 58N	30	CEFAS+SOAEFD	30	RIVO
	England	5/6 to 29/6	43N - 49N	30(W)	CEFAS+SOAEFD	30(W)	RIVO
7	Scotland	29/6 to20/7	44N - 54N	90	CEFAS+SOAEFD	90	RIVO

SOAEFD, Aberdeen, Scotland: Dave Reid
CEFAS, Lowestoft, England: Peter Witthames
IEO, Vigo, Spain: Jose-Ramon Perez
RIVO-DLO, IJmuiden, Netherlands: Guus Eltink
IPIMAR, Lisbon, Portugal: Ana Maria Costa

Table 6.4 Recommended tabulation for maturity at age data at sea and in the laboratory.

Ref. nos inc. cruise (jar label)	Latitude position	Longitude position	Age	Length (cm)	Total weight (g)	Ovary weight (g)	Visual Maturity Stage	Clear visible signs of spawning Y/N	HISTOLOGY			Microscopic Maturity Stage
									Max. viable oocyte diameter (µm)	POFS presence = 1 absense = 0	100% atresia in oocytes > 425µm presence = 1 absence = 0	

Table 6.5   Distribution of ovaries for maturity estimation of mackerel and horse mackerel to be analysed by Laboratory/Country.

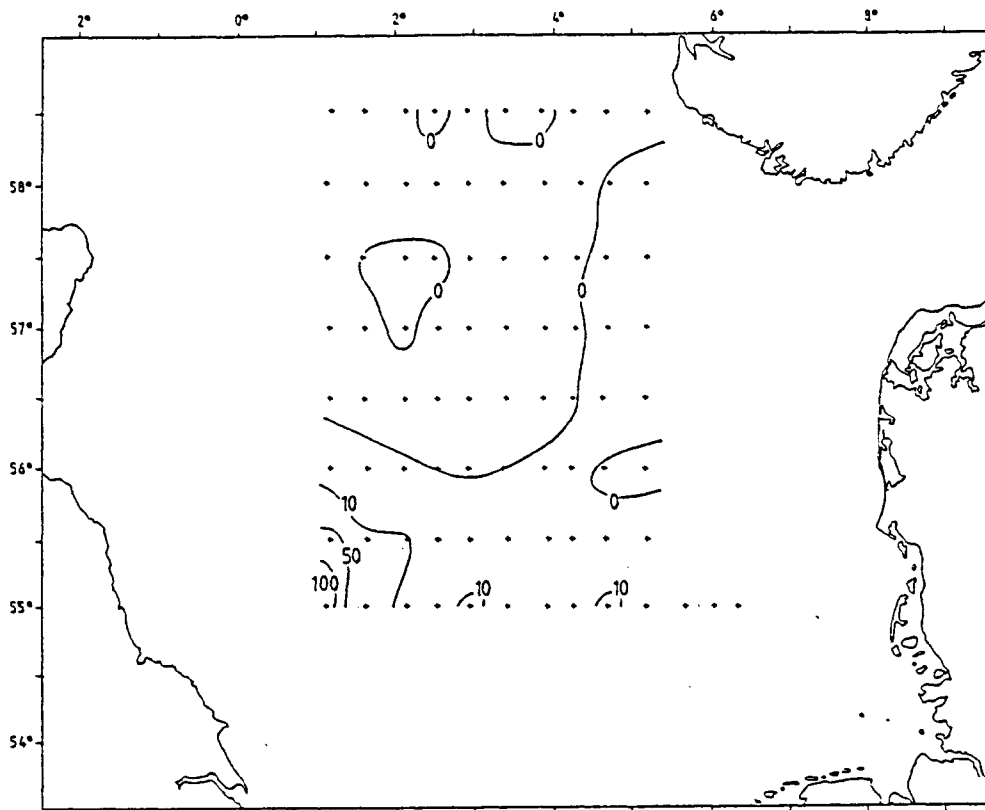| Area	Survey coordinator	Mackerel	Horse mackerel
Western	John Nichols	50% CEFAS 50 % Aberdeen University	50% RIVO-DLO 50% ? FRC
Southern	Fatima Borges	50% IEO Vigo 50% IPIMAR	50% IEO Vigo 50% IPIMAR

Figure 3.1 The distribution of daily production of mackerel eggs per m² during the first coverage, 6–15 June 1996, and the stations sampled.
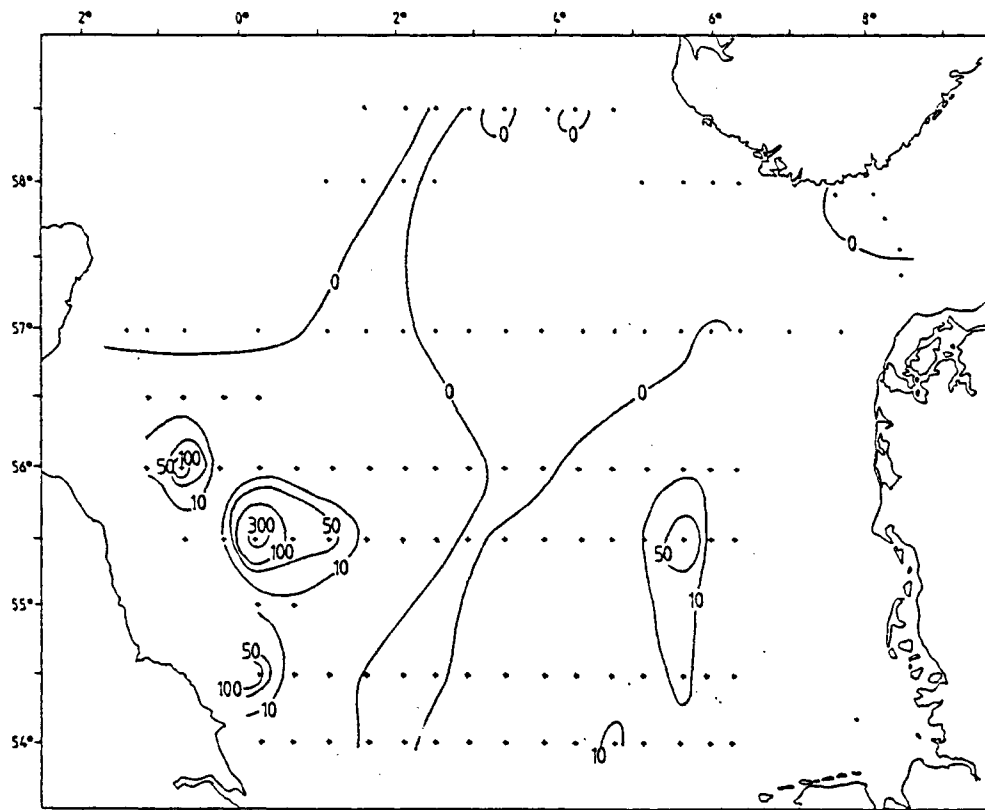

Figure 3.2 The distribution of daily production of mackerel eggs per m² during the second coverage, 15–23 June 1996, and the stations sampled.
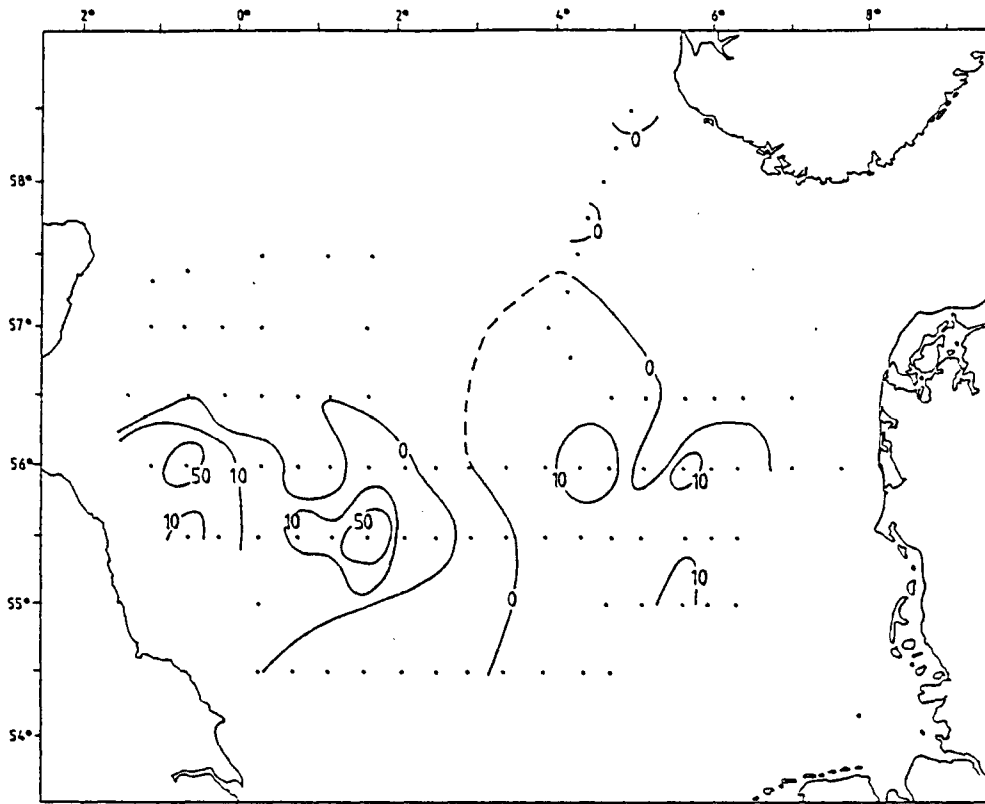
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Figure 3.3 The distribution of daily production of mackerel eggs per m² during the third coverage, 23 June–3 July 1996, and the stations sampled.
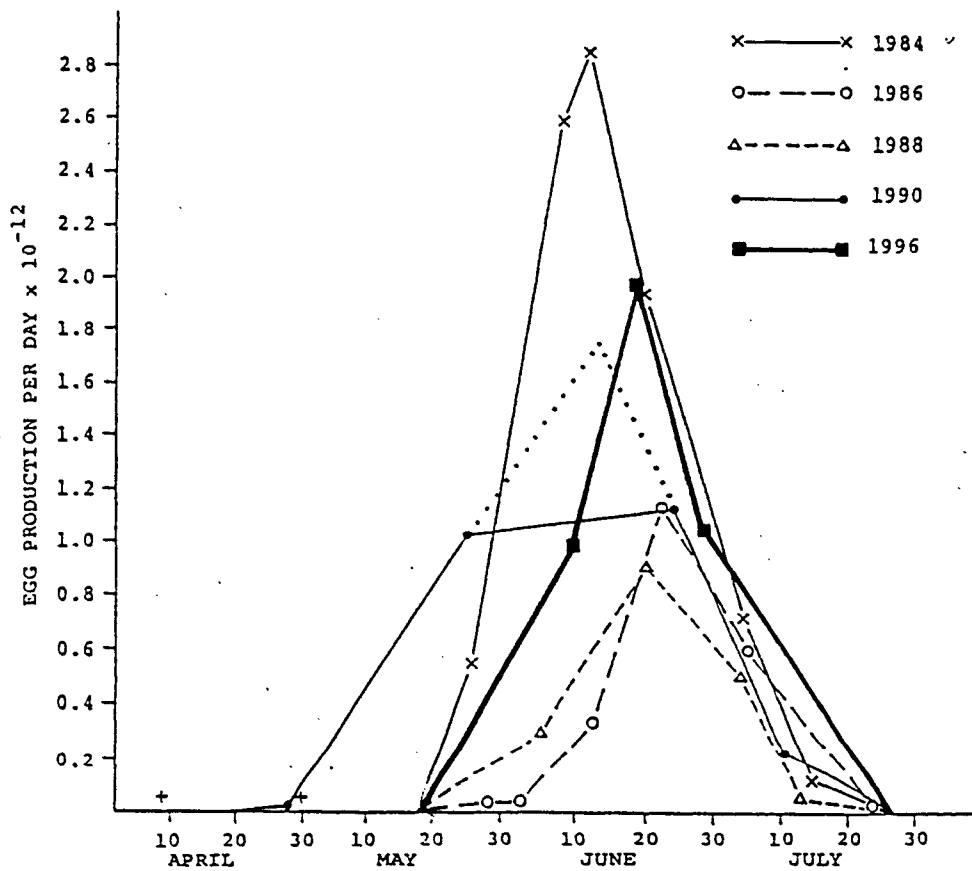

Figure 3.4 Mackerel egg production curves for the period 1984–1996. The + indicates that few eggs were observed during two coverages in April 1988. Dotted line indicates suggested alternative pattern for the peak spawning period in 1990.
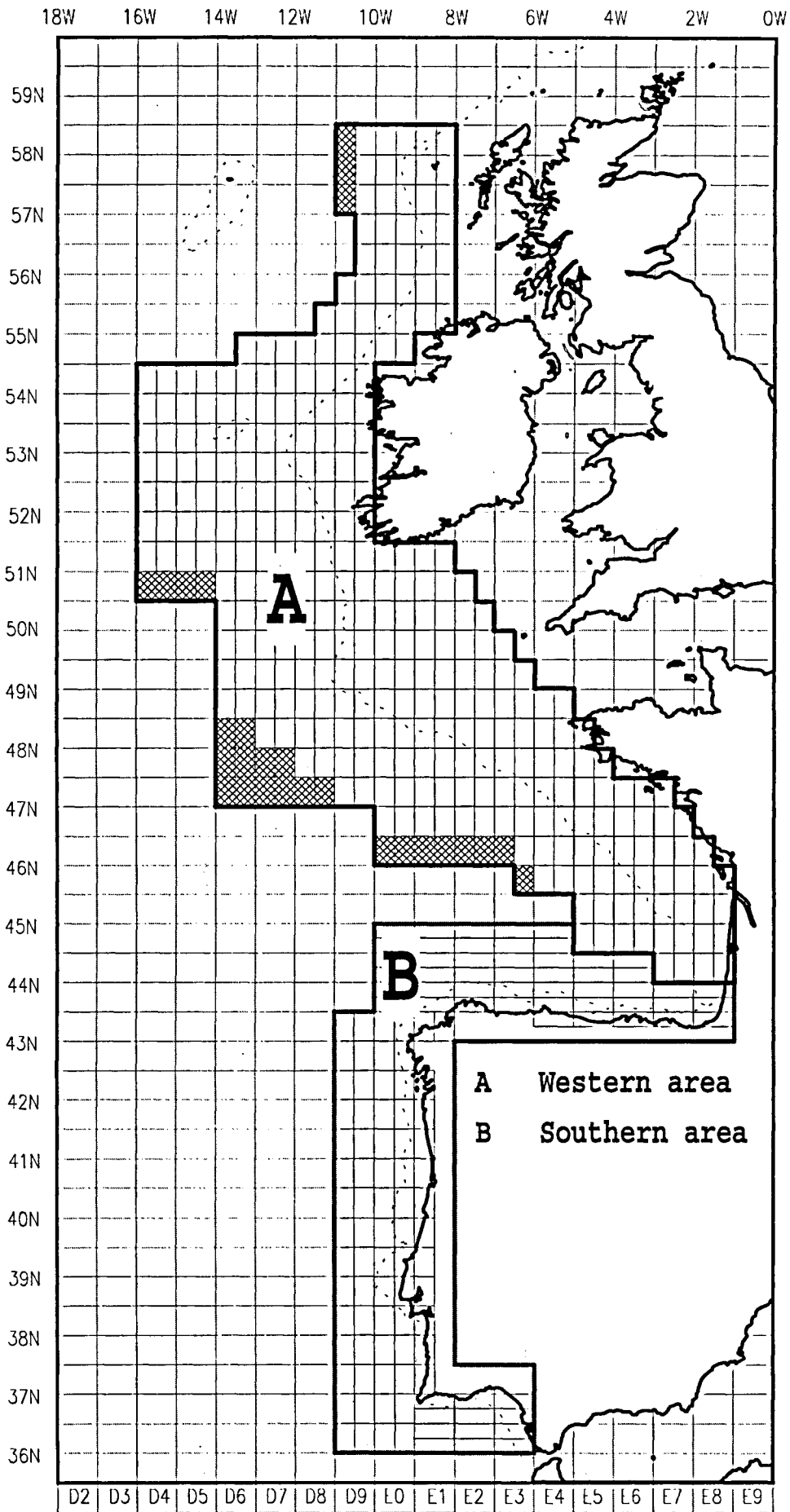
Figure 6.1 Overall sampling area for western/southern components of the mackerel and horse mackerel spawning stock.
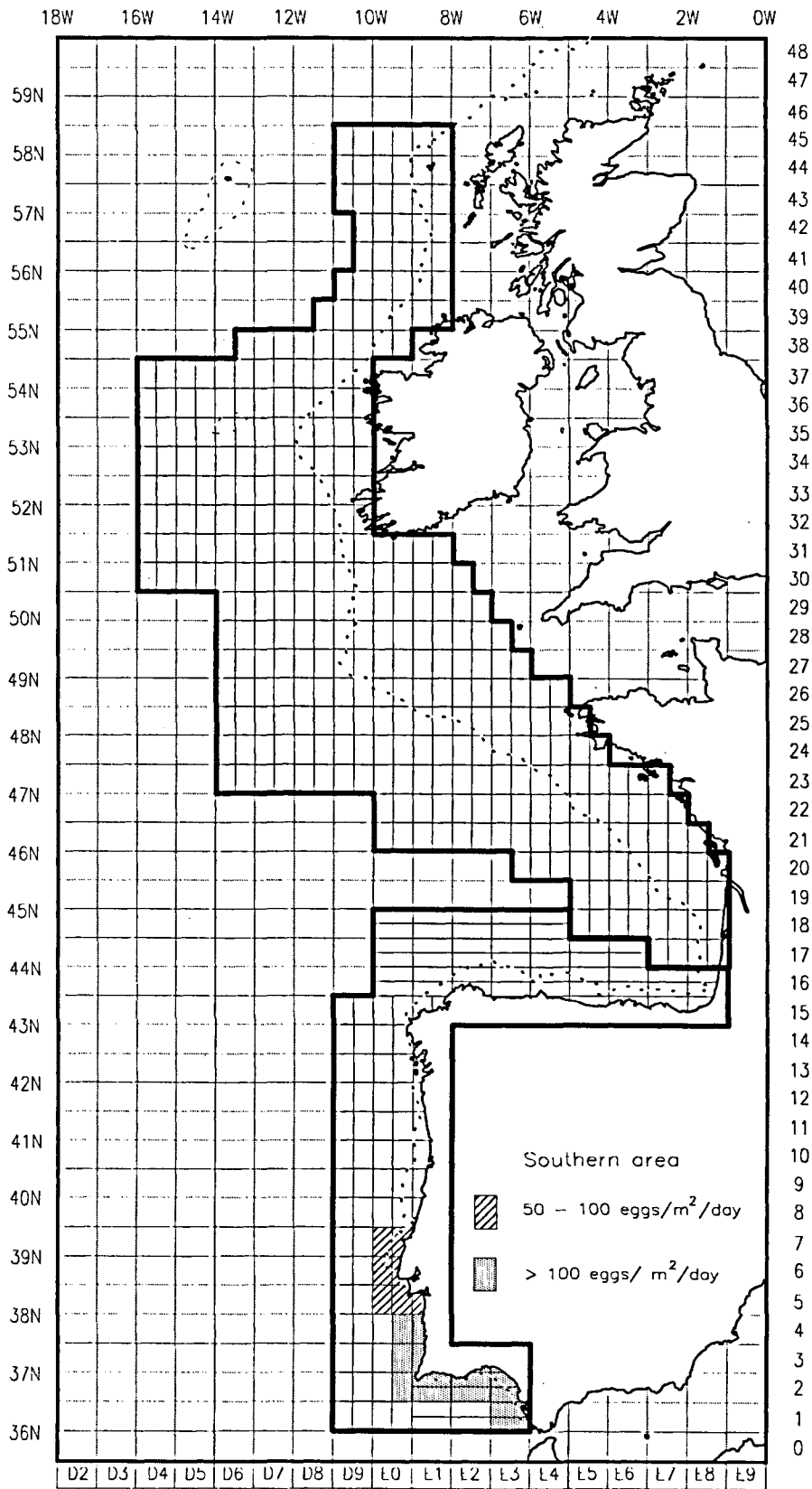
Figure 6.2 Main distributional area of stage 1 Mackerel and Horse mackerel eggs from the 1995 survey. Period 1.
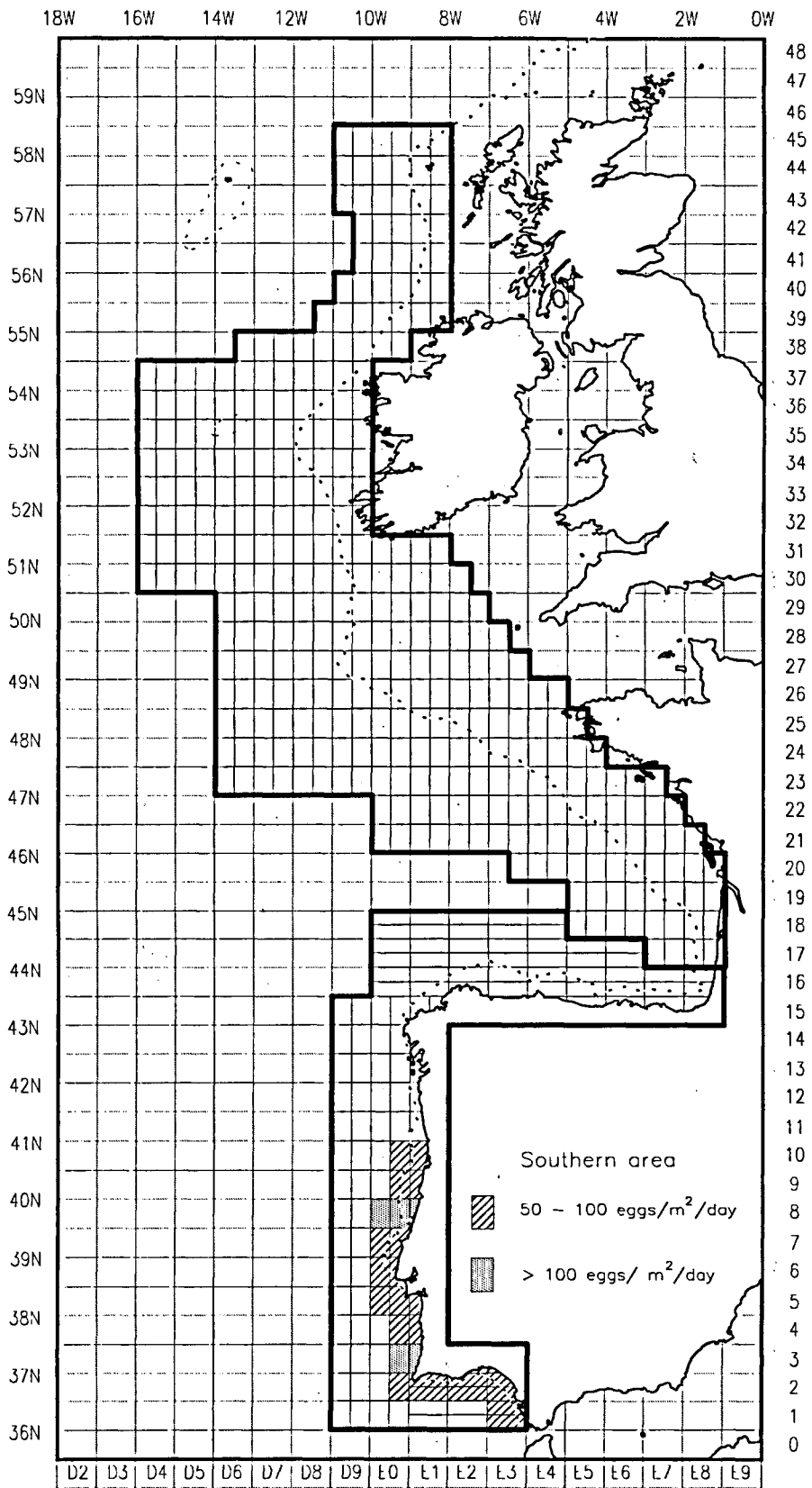
Figure 6.3   Main distributional area of stage 1 Mackerel and Horse mackerel eggs from the 1995 survey. Period 2.
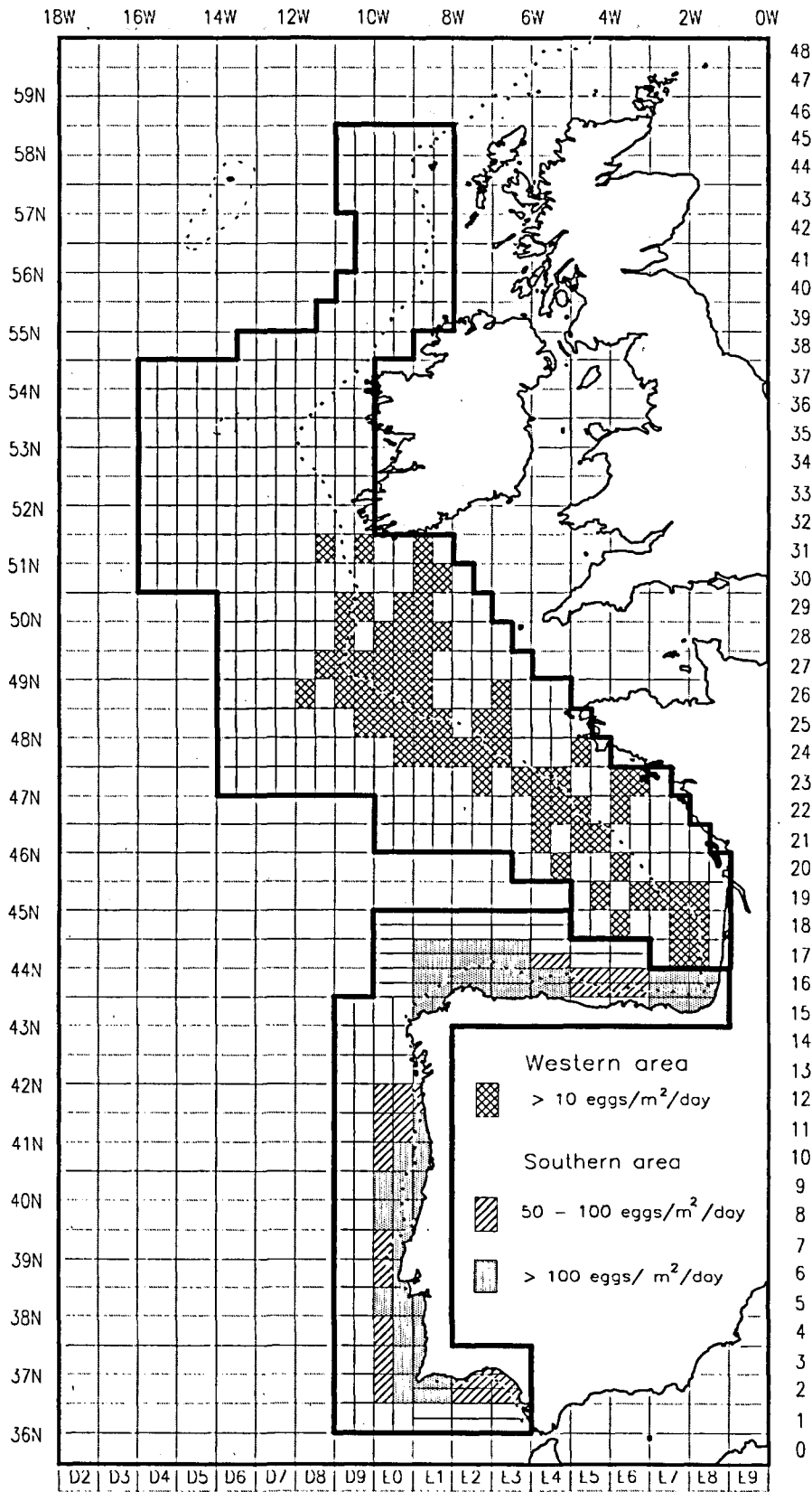
Figure 6.4 Main distributional area of stage 1 Mackerel and Horse mackerel eggs from the 1995 survey in the Western area, 23 March–15 April, and surveys during 1992 and 1988 in the Southern area. Period 3.
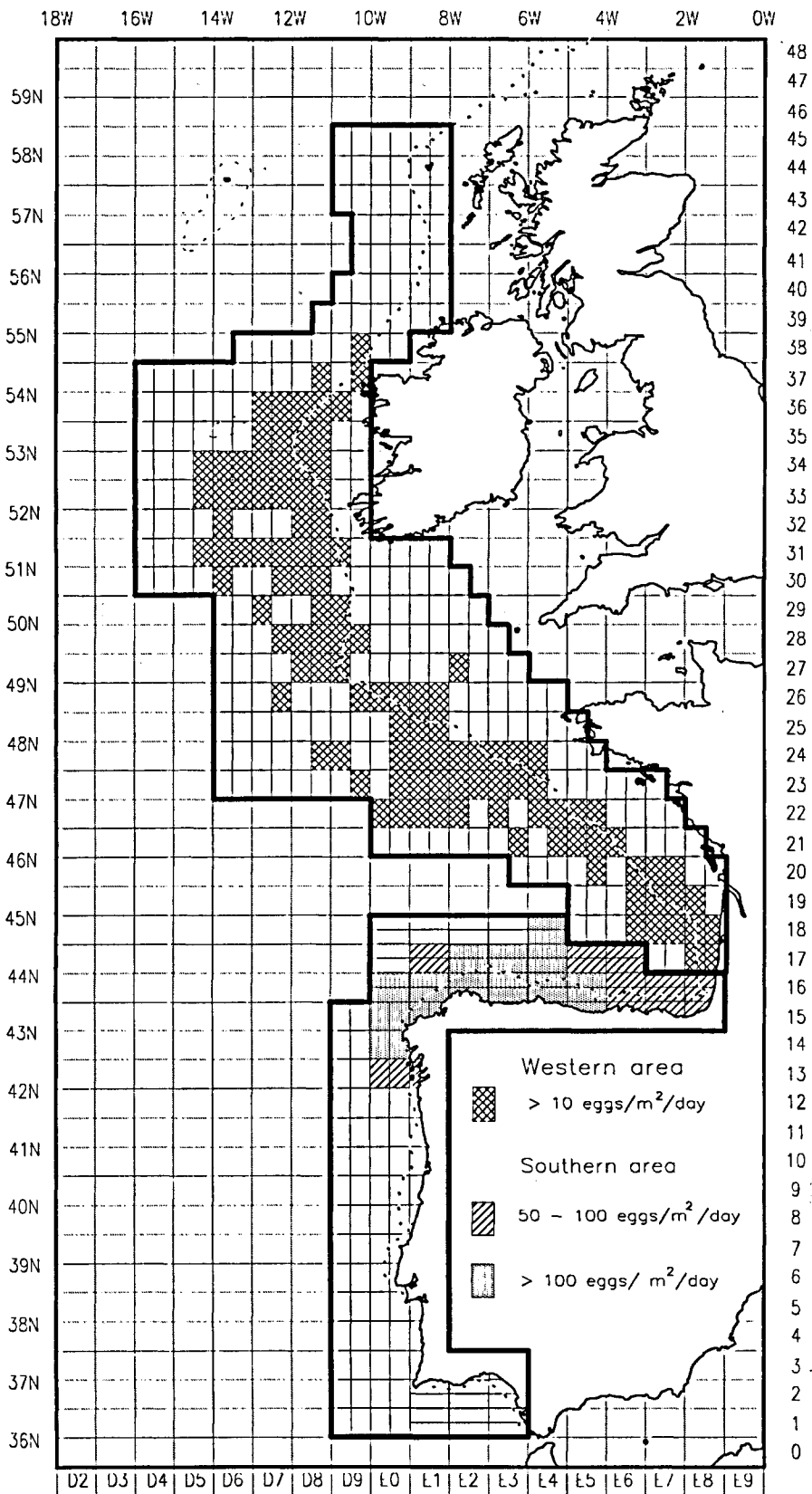
Figure 6.5 Main distributional area of stage 1 Mackerel and Horse mackerel eggs from the 1995 survey in the Western area and surveys during 1990 in the Southern area. Period 4.

Figure 6.6   Main distributional area of stage 1 Mackerel and Horse mackerel eggs from the 1995 survey in the Western area and surveys during 1990 and 1992 in the Southern area. Period 5.

Figure 6.7   Main distributional area of stage 1 Mackerel and Horse mackerel eggs from the 1995 survey in the Western area and surveys during 1992 in the Southern area. Period 6.

Figure 6.8   Main distributional area of stage 1 Mackerel and Horse mackerel eggs from the 1995 survey. Period 7.

APPENDIX 1
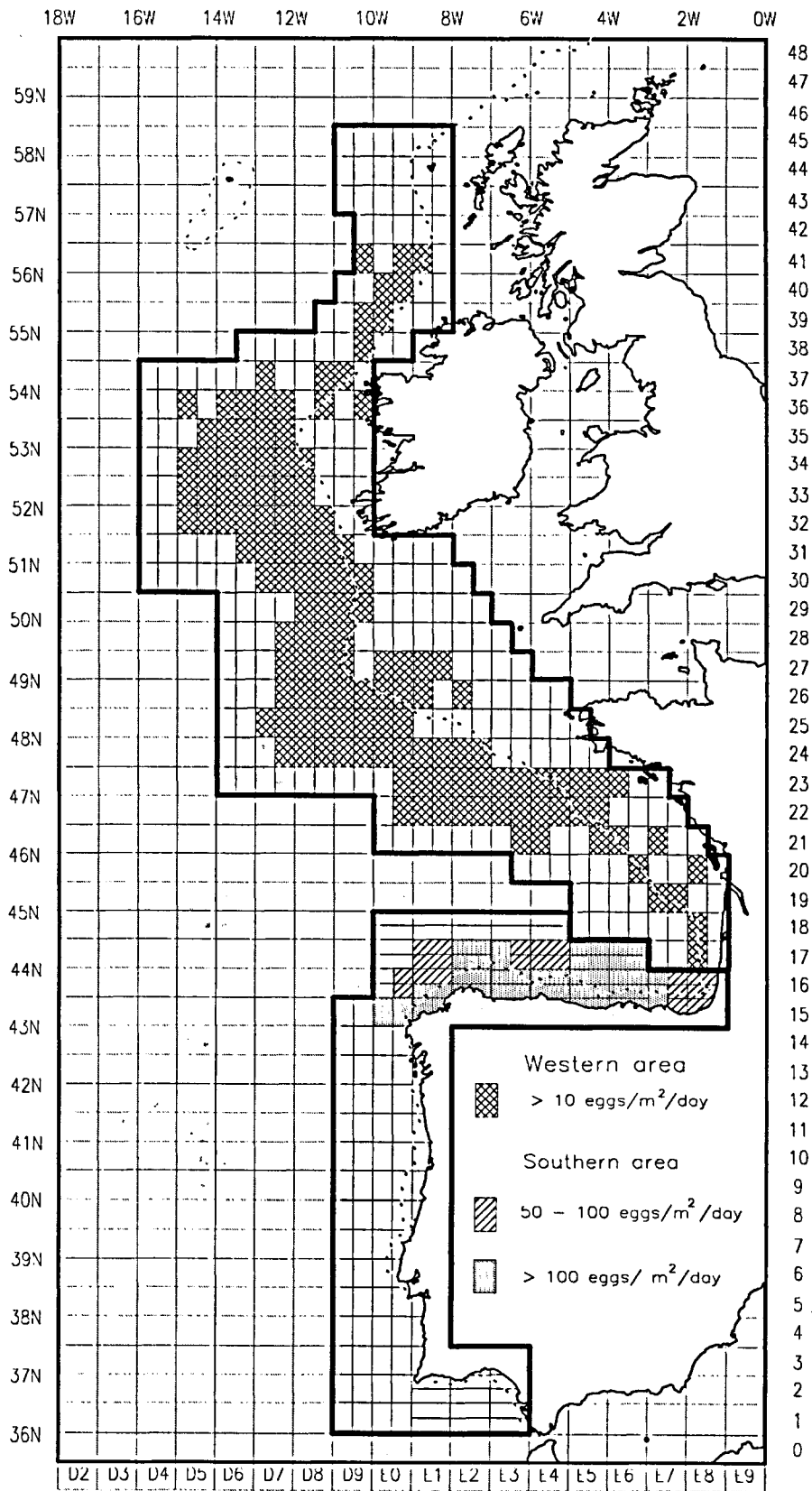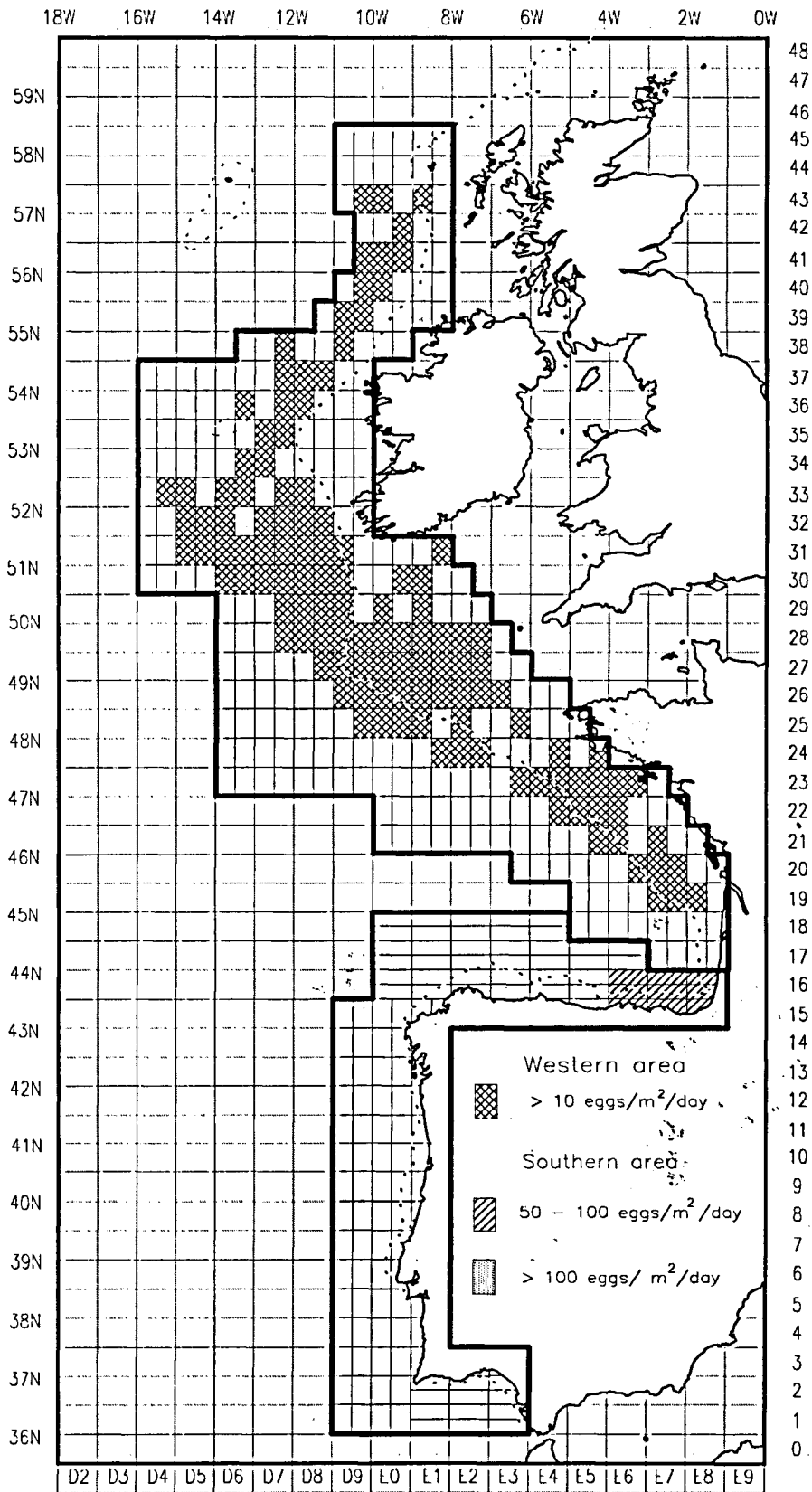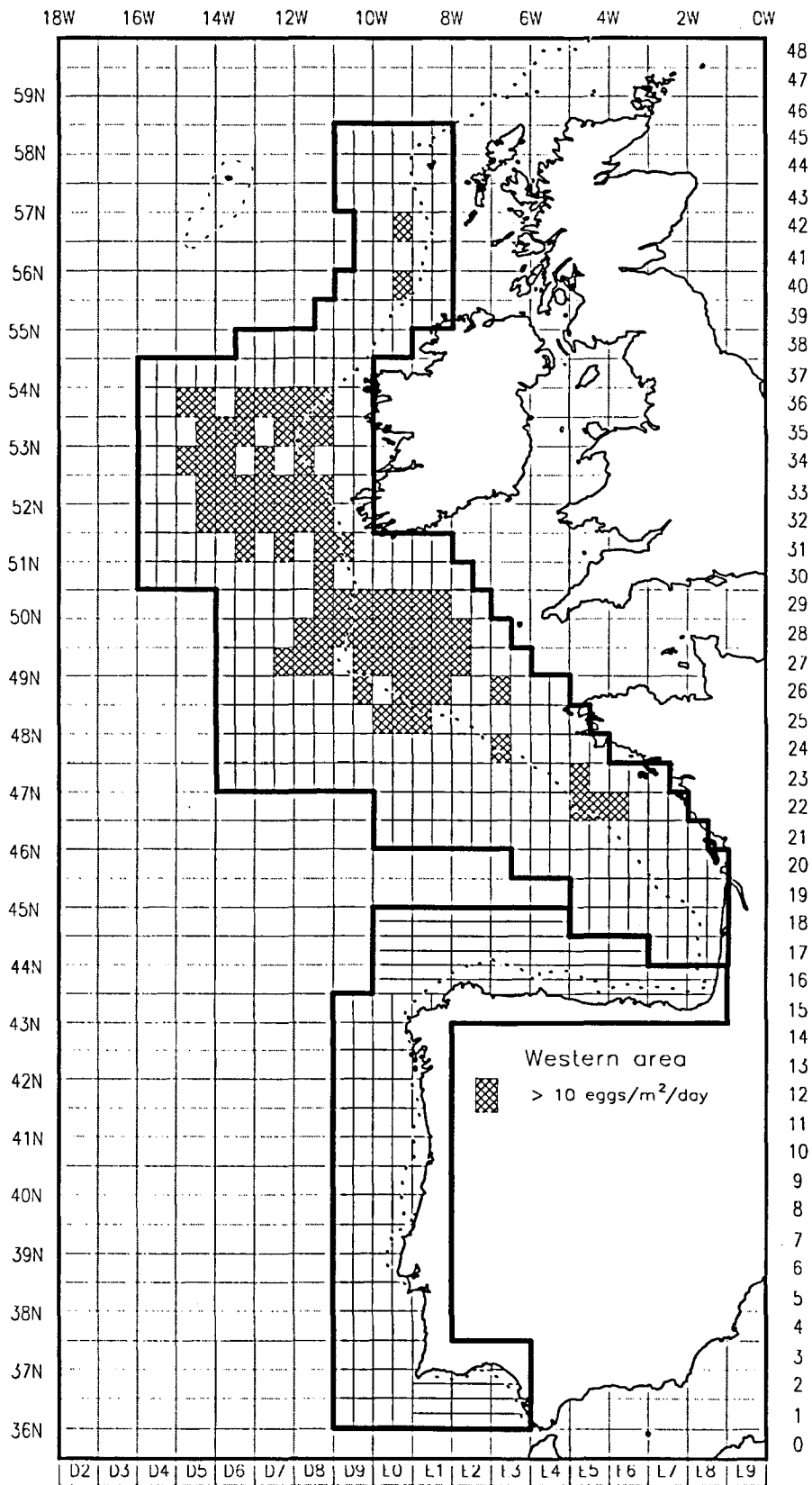
Review of mackerel fecundity and atresia

A review of all the fecundity and atresia data for the Western and Southern areas by Witthames and Maxwell (CEFAS Lowestoft) was presented to the Working Group (WD 1997). Particular reference was made to the significance of inter-annual differences in the values measured.

Each mature female annually produces a stock of eggs (F_{pot} the relative annual potential fecundity measured as eggs g^{-1} total weight) prior to spawning some of which subsequently die in the ovary during spawning (F_{atr} atresia measured as dead eggs g^{-1} total weight) and the remainder which are spawned (F_{real} the realised fecundity) i.e.

$$F_{real} = F_{pot} - F_{atr}$$

The realised fecundity is the denominator below the annual egg production (Egg_{annual}) in the equation to estimate female spawning biomass (B):

$$B = \frac{Egg_{annual}}{(F_{pot} - F_{atr})}$$

F_{pot} was estimated by raising the counts of oocytes $>130\mu m$ found in gravimetric sub samples (Walsh *et al.* 1990) taken from ovaries of females sampled just prior to the start of spawning (March in the Western stock). Atresia is estimated using a sterometric technique Emerson *et al.* 1990) in females at maturity stage 4–6 (Walsh *et al.* 1990) collected at random during the annual spawning season. F_{atr} was estimated in the population using the following equation:

$$F_{atr} = I_{int} \times P_{rev} \times \frac{S}{D}$$

where:
I_{atr} = mean number of dead eggs per fish g^{-1} total weight but excluding fish with no atresia present.
P_{rev} = proportion of fish with atresia.
S = the duration of spawning estimated as 60 days (Dawson 1986, Eltink 1987).
D = the duration of the atretic stage estimated as 7.5 days (Anon., 1993).
Previous results and development of the methods were described and reviewed in Anon., 1990, 1993 & 1996. The data available for all of the triennial surveys is shown in the Table 1 below.

Table 1 A summary of the mean F_{pot} and F_{atr} estimated on the trienniel mackerel egg production surveys since 1977 is shown by the (shaded areas) with the data selected for analysis outlined in bold.

Year	F_{pot} Western area	Southern area	F_{atr} Western area	Southern area
1977	1			
1986	1457			
1989	1608		not calculated 2	
1992	1569		138	
1995	1473	1344	171	

[1] No weight data available for the fish in the fecundity samples.
[2] Data available.

The 1977 and 1986 fecundity data was not considered because no atresia was estimated and no weight infomation was recorded for each fish in 1977. In the case of the 1986 survey F_{pot} was 11% lower than 1989 (Anon., 1990). In this review we consider:

- Analytical methods to estimate F_{atr} in the population.
- A comparison of I_{atr} and P_{rev} estimated by CEFAS and Aberdeen University subcontracted to SOAEFD.
- The temporal distribution of fish sampling to estimate F_{atr} during the 1989, 1992 and 1995 annual egg production surveys.

- Estimation of F_{pot} in Southern (1995 only) and Western stocks in 1989, 1992 and 1995.
- The inter annual differences (1989, 1992 and and 1995) in realised fecundity of the Western stock (the parameters F_{pot}, F_{int} and P used to estimate F_{real} were compared seperately and in combination).

Analytical methods to estimate F_{atr} in the population

The majority of fish do not have atresia (see below). Because of this large number of observations at the minimum value there is not a suitable transformation for the whole dataset. The current approach is using two measures, P_{rev} and I_{atr} for fish with atresia. Applying a log transformation to the I_{atr} for fish with atresia gives an approximately normal distribution. Working with the arithmetic mean (AM) of the logged data is equivalent to using the geometric mean (GM) of the untransformed data as:

GM[data] = exp (AM [log (data)])

A comparison of I_{atr} and P_{rev} estimated in 1995 by CEFAS Lowestoft and Aberdeen University subcontracted to SOAEFD.

A subset of the 1995 western mackerel atresia data was selected from stations with a continuous run of fish samples which had been alternatively sent to either CEFAS or SOAEFD. Therefore other factors affecting atresia intensity do not have to be adjusted for in the comparison. The results of the comparison (Table 2) show that P_{rev} was not significantly different P=0.58 for the two laboratories but I_{atr} (Tables 3 and 4) for fish with atresia has a significantly higher mean and variance at the SOAEFD Laboratory.

Table 2 A   Comparison of P_{rev} results from SOAEFD and CEFAS.

	without atresia	with atresia
SOAEFD	53	24
CEFAS	55	23
Chi squared test,		
H_0 : prevalence equal for the two labs, gives: $\chi^2 = 0.0028$, df=1, p=0.958		

Table 3 A   Comparison of log atresia intensity I_{atr} for fish with atresia between laboratories.

	mean	variance
SOAEFD	4.557	1.263
CEFAS	3.614	0.469

F test for variance equality gives:
F = 2.70, numerator df =23, denominator df = 22, p=0.023
Two sample t-test for equality of means when variances are unequal gives:
t = 3.49, df=38.26, p=0.0012

Table 4 A   Comparison of geometric mean atresia intensity (in oocytes g^{-1}) between laboratories.

	geo. mean	95% CI
SOAEFD	95.30	59.4 , 153.0
CEFAS	37.10	27.6 , 49.9

The temporal distribution of fish sampling to estimate atresia during the 1989 1992 and 1995 annual egg production surveys.

The number of fish from each station in 1989 is similar but in 1992 and 1995 the stations are not evenly represented. In 1992 and 1995 the four stations with the largest number of fish account for about 40% of the total number of fish analysed in that year (see Table 5).

Table 5 Details of the number of fish sampled for atresia by station in the years 1992 and 1995.

1992: total no. of fish analysed 236 from 43 stations		1995: total no. of fish analysed 323 26 stations	
Station	% of total fish from this station	Station	% of total fish from this station
122	15.7	1	20.7
7	11.4	285	9.0
15	8.5	6	7.7
16	6.4	284	7.1
other stations	58.0	other stations	61.7

The uneven spread of samples by station may not cause a problem as all the stations are within the spawning area. However, the number of fish sampled is uneven across time in all 3 years and between laboratories in 1995 as shown in Table 3.3 and these time-laboratory variations are different for each year (see Table 6).

Table 6 Details of the number of atresia samples by month and year.

	1989		1992		1995	
	SOAEFD	CEFAS	SOAEFD	CEFAS	SOAEFD	CEFAS
APRIL	4	4	42	42	58	46
MAY	16	27	31	31	56	24
JUNE	23	27	2	1	85	15
JULY	22	23	44	43	34	5

F_{atr} is known to be sensitive to the point of sampling within the spawning cycle (see below) and a difference between the laboratories' 1995 results has been shown. The overall mean for a year will represent more strongly the sampling periods with more evenly spread observations and the between year effects will be more comparable. The analysis testing for differences between years needs to take this into account.

Estimation of F_{pot} in the Southern (1995 only) and Western stocks during 1989, 1992 and 1995.

Western stocks

The F_{pot} data for the Western stocks are the combined results from CEFAS and SOAEFD which have been compared and accepted by previous working groups (Anon., 1990, 1993 & 1996). A log transformation is applied. The transformed data are approximately normally distributed so analysis of variance is used to test for a significant year effect. The analysis shows one clear outlier which has a large effect on the results of the analysis. Its potential fecundity is 566 oocytes g^{-1} while the second smallest potential fecundity in the dataset is 930 oocytes g^{-1}. Details of the calculation leading to the obsevation were investigated but no explanation appeared so it was removed and the analysis repeated. The Anova table (Table 7) shows there is evidence of differences in F_{pot} between the three years.

Table 7 ANOVA table after fitting the model to $\log(F_{pot}) = $ Year$_i$ for $_i = $ 1989, 1992 and 1995 with year as a class variable (case 1) and a continuous variable (case 2)

	df	SS	MS	F	Pr (F)
year (case 1)	2	0.2427	0.1214	3.09	0.047
year (case 2)	1	0.2422	0.2422	6.17	0.013
non linear	1	0.0005	0.0005	0.01	0.913
residual	267	10.4831	0.0393		

Table 8   Fitted values for $\log(F_{pot})$ = Year$_i$ as a class variable for$_i$ = 1989, 1992 and 1995.

	$\log(F_{pot})$ (eggs g^{-1})	s.e.	F_{pot} (eggs g^{-1})	s.e.	95% CI
1989	7.342	0.020	1543	31.0	1483 , 1605
1992	7.303	0.022	1485	33.3	1421 , 1552
1995	7.271	0.020	1437	29.2	1381 , 1496

(The confidence interval was constructed on the log scale then the limits transformed.)

Partitioning the difference between years into a linear and a non linear component shows there is evidence that the changes in F_{pot} can be described by a linear trend. The slope coefficient is -0.0355 s.e. 0.0143 for $\log(F_{pot})$ which converts to an estimated 3.5% decrease in potential fecundity between surveys.

Comparison of Western and Southern stocks

Normal probability plot shows the normality assumption is reasonable for F_{pot} in the Southern and Western stocks. The following equation: F_{pot} = weight + area$_i$ + weight * area$_i$ where i = Southern, Western areas and weight = total fish weight was fitted to the data (Table 9). Fish weight has a significant effect on F_{pot} (P<0.001) but the area effect is not significant once the weight effect has been taken into account.

Table 9   Anova table after fitting the model F_{pot} = weight + area$_i$ + weight * area$_i$ where i = southern, western areas and weight = total fish weight.

	df	SS	MS	F	Pr(F)
weight | area	1	430777	430777	6.45	0.012
area | weight	1	12031	12031	0.18	0.672
area * weight	1	742623	742623	11.11	0.001
residual	193	12896027	66819		

Table 10   Coefficients for the model F_{pot} = weight + area$_i$ + weight * area$_i$ where $_1$ =Western and Southern areas.

	Value	s.e.
intercept	1725	93.0
area (west)[1]	-380	128.4
weight	-0.61	0.15
area (west) * weight[2]	0.93	0.28

area intercept and slope illiased to the value above.

However, the area*weight interaction is highly significant showing the pattern of fecundity with weight is different in the two areas. This is illustrated by the model coefficients (Table 10) and plots of the data and fitted line for the two areas (Appendix Figure 1). The Western mackerel data shows a non significant trend of F_{pot} (in eggs per gram) increasing with weight while the southern mackerel data shows a significant trend of F_{pot} (in eggs per gram) decreasing with weight (see Table 11).

Table 11   Analysis of variance table after fitting the model F_{pot} = fish weight$_a$ where a = Western and southern areas respectively

Area		df	SS	MS	F	Pr(F)
Western	weight	1	127451	127451	1.76	0.188
	residual	93	6743813	72514		
Southern	weight	1	1045949	1045949	17.0	<0.0001
	residual	100	6152214	61522		

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The negative slope in the Southern area is unusual and the explanation may lie in a failure to detect the start of spawning particularly in the larger fish. This possibility must be investigated by a reexamination of the slides used to check that these fish were caught just prior to the start of spawning.

The inter annual differences (1989, 1992 and and 1995) in the realised fecundity (F_{real}) of the Western stock.

P_{rev} and I_{atr}

The presence of atresia is a binary response so is modelled using logistic regression. The log of I_{atr} (in oocytes g^{-1}) for fish with atresia is modelled by linear regression. Factors used in the model are:

laboratory : SOAEFD, CEFAS to adjust for differences between laboratories.
year :1989, 1992, 1995 to test for differences between years.
period : to adjust for atresia varying across the spawning period.
Terms representing ship, capture method, longitude & latitude and station no. are not modelled.

The 1995 report (Anon., 1996) gave measurements by months but cruises mostly ran across months with few observations in the middle of months. Therefore four equal sampling periods were used to group the data as Table 12.

Table 12   Numbers of fish sampled for atresia during 1989, 1992 and 1995 grouped between 4 periods of equivalent duration.

		Period 1 26 March → 23 April		Period 2 24 April → 23 May		Period 3 24 May → 21 June		Period 4 22 June → 20 July	
Year	lab	Total n	atresia present	Total n	atresia present	Total n	atresia present	Total n	atresia present
1989	SOAEFD	0	0	9	7	34	12	22	6
	CEFAS	0	0	17	6	41	11	23	7
1992	SOAEFD	23	2	25	7	25	7	46	17
	CEFAS	23	10	25	7	25	9	44	12
1995	SOAEFD	23	5	40	16	113	29	57	9
	CEFAS	25	7	21	4	24	13	20	5

Annual Averages

Annual averages are calculated by giving equal weighting to the fitted values from each *period-laboratory* combination. These averages and their SEs are calculated on the log scale for intensity and the logistic scale for prevalence then converted back to the original scale.

Prevalence (P_{rev})

The selected model is:

$$\log (P_{rev} / (1 - P_{rev})) = laboratory_i + period_j + year_k + laboratory.period_{ij} + period.year_{jk}$$

where P_{rev} is the probability of a fish having atresia.

Table 13   The analysis of deviance table (terms added sequentially) for the model log (P_{rev} /(1-P_{rev})) = *laboratory$_i$ + period$_j$ + year$_k$ + laboratory.period$_{ij}$ + period.year$_{jk}$* I.

term	df	deviance	p value
laboratory	1	3.898	0.048
period	3	2.307	0.511
year	2	2.368	0.306
laboratory.period	3	10.494	0.015
period.year	5	13.252	0.021

The *year* term does not give a significant change in deviance so there is not significant evidence for a change in P_{rev} between years. The period.year interaction is significant indicating different patterns of P_{rev} within the three years. The fitted values for P_{rev} by year are shown below.

Table 14 Fitted values for (P_{rev}) after fitting the model shown in the Anova on Table 13.

Year	Lab.	Period 1		2		3		4	
		P	SE	P	SE	P	SE	P	SE
1989	SOAEFD	0.219	0.113	0.628	0.113	0.254	0.059	0.241	0.069
	DFR	0.462	0.148	0.432	0.104	0.350	0.064	0.335	0.081
1992	SOAEFD	0.158	0.063	0.359	0.083	0.270	0.069	0.378	0.063
	DFR	0.364	0.092	0.201	0.064	0.370	0.079	0.491	0.067
1995	SOAEFD	0.147	0.060	0.385	0.071	0.289	0.041	0.163	0.043
	DFR	0.345	0.087	0.220	0.069	0.391	0.073	0.236	0.066

The estimated annual averages with SEs and 95% CI converted from the logistic scale are shown in Table 14.

Table 14 Fitted mean values of prevalence for the years 1989, 1992 and 1995.

Year	Prevalence	SE	95% CI
1989	0.357	0.054	0.260 , 0.468
1992	0.314	0.033	0.254-, 0.381
1995	0.262	0.028	0.210 , 0.321

Atresia Intensity (I_{atr})

The selected model is log $(I_{atr}) = laboratory_i + period_j + year_k$ and the results of the analysis of variance analysis for log I_{atr} after fitting the model are shown in Table 15.

Table 15 Analysis of variance table after fitting the model log $(I_{atr}) = laboratory_i + period_j + year_k$

	df	SS	MS	F	p-value
laboratory	1	29.39	29.39	21.43	< 0.001
period	3	18.69	6.23	4.54	0.004
year	2	26.31	13.16	9.59	< 0.001
Residual	211	289.46	1.37		

The *year* term is highly significant so there is very strong evidence that I_{atr} for fish with atresia is different in the three years. The fitted values for I_{atr} by period in each year are given on both the log (Table 16) and original scales (Table 17). The log scale should be used to make comparisons and construct confidence intervals as shown in the mean predicted annual transformed values for each year (Table 18).

Table 16 Predicted values of log scale I_{atr} after fitting the model in Table 15.

Year	Lab.	Period 1 I_{atr}	s.e.	2 log I_{atr}	s.e.	3 log I_{atr}	s.e.	4 log I_{atr}	s.e.
1989	SOAEFD	5.01	0.33	4.87	0.23	5.39	0.21	4.98	0.23
	DFR	4.35	0.31	4.22	0.24	4.74	0.21	4.33	0.23
1992	SOAEFD	4.04	0.29	3.90	0.23	4.42	0.21	4.01	0.19
	DFR	3.38	0.26	3.24	0.23	3.77	0.21	3.36	0.18
1995	SOAEFD	4.43	0.28	4.29	0.20	4.81	0.16	4.40	0.21
	DFR	3.78	0.27	3.64	0.23	4.16	0.19	3.75	0.23

Table 17 Predicted values of I_{atr} after transformation by period after fitting the model in Table 15.

Year	Period Lab	Period 1		2		3		4	
1989	SOAEFD	149.56	4.10	130.13	2.65	219.79	3.10	145.57	2.83
	DFR	77.86	2.74	67.75	1.98	114.42	2.24	75.79	2.01
1992	SOAEFD	56.63	2.20	49.28	1.58	83.23	1.94	55.12	1.40
	DFR	29.48	1.41	25.65	1.15	43.33	1.36	28.70	0.94
1995	SOAEFD	83.88	2.52	72.98	1.71	123.26	1.80	81.64	1.85
	DFR	43.67	1.76	37.99	1.44	64.17	1.55	42.50	1.48

Table 18 Predicted values of I_{atr} by year after fitting the model in Table 15.

	log I_{atr}	se	I_{atr}	se	95% CI
1989	4.736	0.167	114.0	19.05	82.2 , 158.1
1992	3.765	0.128	43.16	5.54	33.6 , 55.5
1995	4.158	0.124	63.92	7.94	50.1 , 81.5

Realised Fecundity

The results for F_{pot}, I_{atr} and P are combined in Table 19 to give an estimated realised fecundity with approximate SE

$$\hat{F}_{real} = \hat{F}_{pot} - \hat{I}_{int} \times \hat{P} \times \frac{S}{D}$$

$$Var[\hat{F}_{real}] \approx Var[\hat{F}_{pot}] + \left(\frac{S}{D}\right)^2 \left\{\hat{P}^2 \, Var[\hat{I}_{int}] + \hat{I}_{int}^2 \, Var[\hat{P}]\right\}$$

Table 19 Annual values of realised fecundity for the years 1989, 1992 and 1995.

Year	Realised fecundity	approx. SE
1989	1217	79.7
1992	1377	37.9
1995	1303	36.5

The SE for 1989 is larger than for 1992 & 1995 because the estimate of I_{atr} and its s.e. are larger in that year. Significant year effects on F_{pot} and I_{atr} for females with atresia were found while P was not significantly different in the three years. The results are plotted in Appendix Figure 2 and illustrates the year effect when the potential fecundity, atresia intensity and prevalence values are combined to give realised fecundity.

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