International Council for the
Exploration of the Sea

C.M. 1994/H:4
Pelagic Fish Committee

REPORT OF THE MACKEREL / HORSE MACKEREL EGG PRODUCTION WORKSHOP

Vigo, 31 January - 4 February 1994

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1 INTRODUCTION

1.1 Terms of Reference

At the ICES Statutory Meeting in Dublin (Ireland) in September 1993 it was decided (C. Res. 1993 / 2:35) that the Mackerel / Horse Mackerel Egg Production Workshop (Chairman: Mr A. Eltink, the Netherlands) will be held at the Instituto Español de Oceanografía in Vigo, Spain from 31 January - 4 February 1994 to:

a) coordinate the timing and planning of the 1995 and 1996 Mackerel / Horse Mackerel Egg Surveys in ICES Sub-areas IV and VI - IX for estimating spawning stock size;

b) evaluate the accuracy and precision of the estimates of spawning stock size from both the annual and daily egg production methods, and advise on the preferred method;

c) undertake a comprehensive review of survey and analytical techniques (consider techniques other than arithmetic averaging for estimating unsampled rectangles and consider how the vertical hauls with a much lower volume filtered have to be treated for the standard error estimation);

d) complete the analysis of the daily egg production method applied to the southern horse mackerel stock based on the 1992 egg and trawl survey data.

1.2 Participation

The Workshop met in Vigo from 31 January - 4 February 1994 with the following participants:

Nicolas Bez	France
Fatima Borges	Portugal
Stephen Buckland	UK (Scotland)
Pablo Carrera	Spain
Guus Eltink (Chairman)	Netherlands
Anabela Farinha	Portugal
Alberto Garcia	Spain
Philippe Guiblin	France
Svein Iversen	Norway
Paulino Lucio	Spain
Lorenzo Motos	Spain
John Nichols	UK (England and Wales)
José Ramón Pérez	Spain
Carmela Porteiro	Spain
Monty Priede	UK (Scotland)
Amor Solá	Spain
Karl-Johan Stæhr	Denmark
Luis Valdés	Spain
Begoña Villamor	Spain
Martin Walsh	UK (Scotland)

2 GENERAL ASPECTS

2.1 Comparison of Egg Staging

Sample exchanges between participating countries, to compare horse mackerel egg staging, began in 1986 and were reported in Anon. (1987). Seven countries participated and the results showed that there was a good agreement on the stage I eggs but that difficulties arose in staging older eggs. The variation from the overall mean stage I count was +19% to -12%.

A further exchange of mackerel eggs took place between six countries in 1989 (Anon., 1990). Once again there was a wide variation in the identification of some stages including stages Ia and Ib. However, most importantly, stage I (Ia + Ib) were fairly accurately identified by all countries with a variation of +10% to -7% from the overall mean, with a coefficient of variation (CV) of 9%. The CV for all other stages ranged from 23% to 81%. A similar exchange of egg samples in 1992 included horse mackerel as well as mackerel and the preliminary results, which excluded Netherlands and Spain 1 and 2, were reported in Anon. (1993a). The final results of the counts of all stages are shown in Table 2.1a for mackerel and 2.1b for horse mackerel. The individual coefficients of variation for each species are shown in Table 2.1c for mackerel and 2.1d for horse mackerel. As in previous exercises the results show a wide variation in the identification of the stages of mackerel eggs. This also now applies to horse mackerel eggs. However the variation between countries is less for the first and last stages than for the intermediate stages. The coefficient of variation for stage I mackerel eggs was 8.8% and for horse mackerel 9.6%. Only stage I eggs are used for spawning stock biomass estimation.

The egg exchange exercises to date have only been used as part of an on-going training programme in an attempt to improve the precision of staging. The data have never been used to modify the counting of egg stages by any country.

The Workshop recommends that a further egg exchange exercise designed to improve the precision of staging will be organised in 1995 by UK (England).

2.2 Egg Stage Duration

Experiments were conducted in 1993 to observe the rate of development of horse mackerel eggs through the blastula stage (Motos & Muriel, WD 1994) following a similar approach to that of Nichols & Warnes (1993) for mackerel. In addition, observations were also made on the duration of developmental stages through to hatching. These observations were made at 5 temperatures from 10°C to 20°C. The text table below gives the coefficients of the fitted equations $Lnt = A\ LnT + B$ where t is time in hours to the end stage I and T is temperature.

Stage	Coefficients		R^2
	B	A	
I	8.153	-1.721	0.94

The results of these additional observations are compared with the calculated durations of these stages using the relationship between development time and temperature given by Pipe & Walker (1987), in Figure 2.1. When comparing these results, the equations of temperature dependent developmental rates are more similar in the higher range of temperature than at the lower temperature. For all the stages fitted, the equations show a slower developmental rate in the 1993 experiment. This reduction is greater as the temperature decreases. This retarding of development could be the effect of the low salinity waters (32 to 34ppt) used in the experiment (Alderdice & Forrester, 1971). In addition low salinity also makes the eggs sink and accumulate in the bottom of the incubation tubes, provoking a shortage of oxygen which is likely to retard development even more (Walsh *et al.*, 1991).

The 1993 study presented non-optimal experimental conditions. As a consequence hatching did not occur at all incubation temperatures and development did not progress beyond Ia stage at 10°C. Within the temperature range where eggs did hatch, the results obtained were similar to those of Pipe & Walker (1987). Given those facts, the workshop believes that the equations of Pipe & Walker should continue to be used to transform stage I egg abundances to daily egg production rates. It remains a possibility that the temperature experienced by individual spawning horse mackerel may affect egg developmental rates (Jennings & Pawson, 1991). In order to test this, a more extensive experiment than the one described here would have to be conducted.

The equations from the early egg stage data set were used to estimate diel spawning time from field data. Field egg samples were collected during DEPM surveys carried out in spring (May-June) from 1989 to 1992 along the Bay of Biscay. All the horse mackerel eggs caught in these surveys were examined and the early blastula stage eggs were identified and counted. They were classified into 5 sub-stages: undifferentiated/1 cell, 2 cell, 4 cell, 8 cell and 16 cell. Egg counts per haul were converted to concentration as numbers per square meter. Each stage (2-16 cells) was then aged by applying the temperature development equation from Motos & Muriel (WD 1994) using the sea surface temperature at each station. Diel spawning time was estimated for each early stage observed in the field by subtracting its estimated age from the collection time of the sample. First cleavage was assumed to occur 30 minutes after spawning. In this way, 228 observations of diel spawning time were obtained.

Figure 2.2 shows the diel spawning pattern of horse mackerel by plotting the incidence of early e.g. occurrences versus back-calculated spawning time. Horse mackerel mainly spawns in the second part of the day, from 14.00 to 20.00 hours (UTC). Very little spawning was observed during the morning, from 4.00 to 12.00 hours (UTC). At night, spawning continues at lower rates and a secondary spawning peak is apparent between 2.00 and 4.00 hours (UTC).

The fact that horse mackerel shows a definite diel spawning cycle is relevant when applying the DEPM to this species. Spawning fraction of horse mackerel females could be estimated by sampling in a period prior to or after spawning using the incidence of identifiable gonadal stages e.g.: nuclear, migration, hydration, early post-ovulatory follicles.

An apparent secondary peak of spawning appeared at night. However, most samples with early egg stages appearing in that period were collected in 1992. Information from adult gonadal stages from the horse mackerel DEPM survey (Anon., 1993a) showed a consistent prevalence of early post-ovulatory follicles at a similar period of the night. It is possible that this secondary peak of spawning at night in 1992 was unusual.

2.3 State of Research on the Duration of Oocyte Development Stages and of Early Post-Ovulatory Follicles

For the purposes of the DEPM an estimate is required of the fraction of spawning females in the spawning stock. The basis of this measurement is to examine a sample of ovaries histologically and identify the fraction of the population that contain oocytes at a stage close to spawning. The spawning fraction S is then given by the equation:

$$S = S_o.24/t_o$$

where S_o is the fraction of fish with hydrated stage "o" oocytes and t_o is the duration of stage "o" in hours. In practice, any of the oocyte development stages close to spawning can be used as an indicator of spawning fraction, the migratory nucleus stage S_{mns}, hydrated stage S_h or post-ovulatory follicle S_{pof}. The stage chosen should have a duration not greater than 24h and not so short that their occurrence in the population is low.

In previous applications of the DEPM to mackerel and horse mackerel in 1989 and 1992 it was assumed that t_{mns} is 24h and therefore:

$$S = S_{mns}$$

This assumption has never been fully verified and hence previous workshops have recommended that further research is required to examine the duration of these stages.

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Spawning has been observed in captive populations of mackerel held in tanks at the Marine Laboratory in Aberdeen. Several times distinct peaks of egg production occurred every 24h which could be generated by different fish spawning each night but the simplest hypothesis

would be individuals spawning at 24h intervals. In histological sections of ovaries of actively spawning fish the hydrated oocytes of a current batch are visible together with the migratory nucleus stage oocytes of the subsequent batch. If it is accepted that mackerel can spawn at approximately 24h intervals this suggests a minimum migratory nucleus stage (MNS) duration of about 24h. If the stage were longer, overlapping generations would mean that the number of migratory nucleus oocytes would exceed the batch fecundity (Priede & Watson, 1993). Some indication of the duration of the MNS duration was also derived from the relative prevalence of the different oocyte stages in the 1989 trawl survey. The prevalence of the different spawning states in a randomly sampled population should be in direct proportion to the stage durations. In 1989, for the western stock, the ratios were: migratory nucleus stage - 3.2, hydrated stage - 1 , early post-ovulatory follicles - 0.6 and late post ovulatory follicles 2.9. In most fishes the hydrated state is an unstable state and in the anchovy begins about 12h before spawning (Hunter *et al.*, 1985). If the duration in mackerel is similar, this suggested a duration for the migratory nucleus stage of not more than 38h.

In the previous egg production workshop (Anon., 1993a) data were presented indicating that in captive fish under stress the MNS could persist for 9-11 days. It was recommended that trawl samples from the 1992 survey in which spawning fraction was 100% should be re-examined to determine whether the number of MNS oocytes exceeded the batch fecundity. If the MNS duration is equal to minimum spawning interval (24h) then the number of MNS oocytes should be equal to the batch fecundity. Three trawl hauls were found with 100% spawning fraction. Both batch fecundity (hydrated oocytes) and number of MNS oocytes were measured for 34 fish. Batch fecundity was 62.53 oocytes g^{-1} (SD = 38.31) and MNS number 63.49 g^{-1} (SD = 27.77) indicating a MNS duration of 24h. Relative prevalence data indicate a hydrated stage duration of 13.9h (Diack & Priede, WD1994). Examination of field data therefore gives no indication that the assumption of 24h duration for the MNS stage should be modified.

In 1993 mackerel were again kept under observation in tanks in the Marine Laboratory, Aberdeen during the spawning season. None of these fish spawned. Therefore no further progress was made on mackerel. Further experiments are planned in 1994.

HORSE MACKEREL

In 1990, 1991 first experiments were conducted by AZTI/SIO using live-bait holding tanks on board the "Divino Jesus de Praga" a chartered commercial tuna fishing vessel. Freshly caught horse mackerel were maintained in sea water for observations on spawning for up to 4 days.

In 1993 with the aid of funding from the EC, AZTI/SIO has established a holding facility for horse mackerel comprising 4 large circular fish tanks together with a recirculatory system controlling, salinity, temperature and general water quality to simulate oceanic conditions. This has for the first time enabled observations of spawning in captive populations of horse mackerel held on shore (Lucio, WD 1994).

In 1993 on two occasions live fish were captured using the fishing vessel "Siempre Ongi Etorri" which were transferred to the newly built holding facility at Sukarrieta.

Ship board observations 1990, 1991

In these experiments it was observed that spawning females could be clearly identified by the presence of a swollen abdomen and extrusion of eggs with a very light pressure applied anterior to the vent. Such fish were marked with individually identifiable external tags and they were observed for subsequent releases of batches of eggs. A variable fraction of the fish would develop this running state at intervals post-capture from a few hours up to several days. Fish were sacrificed for examination of the ovaries at varying intervals after the onset of the swollen state. Hydrated oocytes were observed up to 31 hours after swelling of the abdomen. This suggests that the hydrated stage may have a much longer duration than the 6-12h typically observed in pelagic fishes. The possibility that this is an effect of stress following capture cannot be excluded.

In post-mortem histological analysis ovaries were seen with 3 distinct stages of post-ovulatory follicles (POFs) indicating spawning of a recent series of batches.

Shore based observations 1993

In the first experiment a population with 104 females was kept under observation and in the second experiment 231 females were observed. 14 and 20 females developed into the swollen spawning state in the first and second experiments respectively. Males with running milt could also be recognised in these tanks. The spawning state in females persisted for up to 28h before they were sacrificed for histology suggesting again a long duration for the hydrated stage.

Selected individually marked spawning females were transferred to a smaller tank with a group of running males. Continuous egg production was observed for a period of 3-5 days with a peak at 2-3 days.

Data from these experiments have not been fully analyzed and it would be premature to draw firm conclusions. Taking into account the low spawning fraction of horse mackerel in the western stock (circa 10%) indicating a batch interval of 10 days it seems that fish may take several days to release a single batch in a series of pulses of eggs. The results of captive spawning experiments may not be representative of events in free-living fishes.

Analysis of data will continue at AZTI/SIO during 1994. So far these experiments have not resulted in an estimate of the duration of any of the oocyte or post-ovulatory follicle stages used for determining spawning fraction.

2.4 Long-term Changes in Batch Fecundity in Mackerel

If batch fecundity could be assumed to be constant from year to year it may be possible to reduce the cost of the

adult fish survey part of the DEPM. To further examine changes in batch fecundity in the western mackerel stock samples were taken by the RV Corystes using a Swedish type "FOTØ" trawl from the following stations (Watson *et al.*, WD 1994):

Station 01:	50°00'N	11°00'W	17 June 1993
Station 41:	51°12'N	10°45'W	24 June 1993
Station 48:	49°55'N	10°36'W	24 June 1993

Batch fecundity was counted for 52 pairs of ovaries according to the methods of Watson *et al.* (1992). These data were compared with archived data from previous years from the same area. All available samples between 49°-51°N and 9°-11°W were included:

Year	n	Dates
1989	54	23 May - 12 June
1991	56	27 May - 12 June
1992	64	25 May - 12 June
1993	52	17 June - 24 June

The linear regression was fitted (forced through zero) to the relationship between batch fecundity and total wet body weight. The slope of the line is equal to F_{bw}, the fecundity per gramme body weight:

YEAR	F_{bw}	r	P
1989	66.61	0.563	<0.001
1991	61.40	0.391	<0.01
1992	51.6	0.542	<0.001
1993	51.8	0.389	<0.01

There appears to be a significant decrease in F_{bw} over time. These batch fecundities in this sampling box (49°-51°N, 9°-11°W) are higher than the average for the whole stock. Overall F_{bw} was 53.05 g^{-1} in 1989 and 46.19 g^{-1} in 1992. Since batch fecundity varies in both time and space within a spawning season the trend may not indicate an absolute decline in batch fecundity from year to year. The apparent downward trend could be the result of change in timing of peak of spawning. The results emphasise that if the DEPM is applied to a stock the batch fecundity should be determined each time, and at the same time as the plankton survey, as recommended by Hunter *et al.* (1985).

This sampling series can be continued in future. Any vessels operating in the 49° - 51°N, 9° - 11°W box during the spawning season are requested to send mackerel ovary samples to the University of Aberdeen.

2.5 Spawning at Different Depth Strata

Mid-water trawls have been used to sample mackerel to estimate both spawning fraction and atresia whilst rod and line have only been used to collect atresia fish samples as part of the method to estimate female spawning stock biomass (Anon., 1990; Anon., 1993a). However, anecdotal evidence in 1988 and 1989 suggested that trawl catches, made below 40 m with a large pelagic trawl, had a lower proportion of running females than samples obtained with rod and line which were usually taken above 20m depth. To investigate whether sampling depth may bias estimates of spawning fraction and atresia the vertical distribution of spawning mackerel was investigated by mid water trawl and rod and line.

Conclusions (Anon., 1993a) from a preliminary analysis to investigate the vertical distribution of mackerel using a mid-water trawl are presented in Table 2.2. Mid-water trawl catches of mackerel during the day from the surface layer (5-15 m depth) were approximately 4 times greater compared to those taken at 50-60 m depth and 10 times greater than at 100-110m. The only observations made around mid night suggested the population was very much aggregated near the surface with catches decreasing rapidly from 50 to a 100 m. Histological analysis of ovaries taken from a sub-sample of these fish suggested that a higher proportion of fish were spawning in the surface layer down to 50 m. These results were supported by a report at the same meeting on the vertical distribution of mackerel eggs, in the plankton during June. In this period the concentration of up to 16 cell stage eggs above 50 m in depth indicated most of the spawning was near the surface.

During 1993 further investigation of the mid-water distribution of mackerel and horse mackerel was undertaken using the same method as described in Anon. (1993a), except that the trawl was deployed from RV Corystes rather than RV Cirolana. The cruise dates were almost two weeks later commencing work at the end of the third week in June on the Great Sole Bank around 49°75'N and 10°30'W with just under 200 metres depth of water in the survey area. Maturity stages were assessed following the reclassification of maturity stages in Anon. (1990). A series of three trawls of one hour duration each was made with the head-line a 0, 50 and 100 metres. Each series of hauls was centred around one of three periods in the 24 hour cycle mid-day, mid-night and just before dusk.

Spawning fish (stage 4 and running fish) were found to comprise a large proportion (41%) of the mature females caught. The mean catch weights of mackerel and horse mackerel at each depth are shown in Table 2.3. Unfortunately very few fish were caught in daylight hours in marked contrast to the night series when both horse mackerel and mackerel were caught in substantial numbers. Around 20.00 hours UTC (dusk) the mackerel catches improved with many more fish in the surface trawls though horse mackerel appeared more uniformly distributed. Around mid-night the mean catch was >10 times higher for both species at the surface compared to 50 metres and >25 times higher compared to 100 metres.

The low catches during daylight hours at all depths trawled compared to the previous observations were quite unexpected. Acoustic information did not suggest any concentration of fish outside of the three trawl depths, for example close to the ocean bed, and generally the population seemed more dispersed and patchy because of the mixed success in obtaining samples using rod and line and the low abundance of early stage mackerel eggs in ring net samples.

The concentration of fish near the surface at night was consistent with the decline in catches during the hours of darkness (765 kg day and 53 kg night) reported from the Kings Cross on the 1992 survey (Anon., 1993a). Previously it was concluded that 'circadian behavioural

changes make mackerel less catchable to pelagic trawl at night' but the explanation maybe because a large commercial trawl, as used by the Kings Cross, under samples the upper 40m of the water column. The high catches of mackerel which appear to be concentrated near the surface at night would suggest that a more representative sample for population atresia measurements should preferably be caught with a mid water trawl avoiding hours of darkness. To keep the headline close to the surface, as with the FOTØ trawl, it would require floats attached to the wing ends and to deploy the trawl at speeds above 5 knots with over 400m of warp.

2.6 Atresia

A further atresia experiment on mackerel was carried out in 1993. Analysis of the data is not yet complete, but should be available by the end of 1994. On completion of the sample analysis, and after combining all the results, some reappraisal of the report in Anon. (1993a) may be necessary.

2.7 Duration of Spawning of Individual Female Fish

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Mackerel spawn the oocytes which mature in their ovary each year (mean annual realised fecundity of about 450,000) as a series of batches with each on average comprising approximately 14,500 oocytes (Anon., 1993a). The duration of spawning of an individual fish maybe defined as:

$$D_S = B_n * B_i$$

where D_S = The duration in days the average fish will take from commencing spawning to spent.
B_n = The average number of batches produced each year per individual female.
B_i = The average time interval for an individual between producing a batch of eggs.

The spawning duration D_S has been used as a parameter to correct the potential annual fecundity of sole for degeneration of yolk oocytes through atresia (Horwood, 1992).

$$F_p = F_r - \frac{A * D_S}{D_a}$$

where F_p = Potential specific fecundity oocytes g^{-1}
F_r = Realised specific fecundity oocytes g^{-1}
A = The instantaneous loss of oocytes from the ovary identified as the prevalence and intensity of atretic oocytes in ovaries sampled over the whole annual egg production cycle.
D_a = The duration oocytes identified in A remain in the ovary.

In the 1992 triennial survey the potential fecundity was reduced by 7.5% (Anon., 1993a) after provisionally assuming a duration of D_S to be 2 months. Any error in this assumption would produce a proportional effect on the atresia correction applied to the final spawning stock biomass estimate. This report reviews the historical data available to estimate D_S and presents some new data from the 1992 triennial egg production survey.

The maximum annual spawning duration could be taken from the number of days at the base of the egg production curve. In the 1992 triennial survey back calculating the age of each egg found in ichthyoplankton samples predicts spawning started on 27 March. The last stage 1 egg was found on the 11 July giving a spawning duration 106 days. However Dawson (1986) and Eltink (1987) describe changes in the size-age distribution of mackerel in the Western Atlantic population which show that individuals reside on the spawning grounds for considerably less than 106 days. For example Eltink (1987) describes a slow change in the size of spawning fish from March to May followed by a sharp decrease from June. The rapid decrease in size was caused most probably by the emigration of April to May spawning fish as spent fish in June. The spawning duration would thus be about two months.

Further data supporting this conclusion can be seen in the appearance of spent fish in the spawning populations sampled for atresia during the 1992 triennial survey. Random fish samples were collected over a wide area at 5 periods in the egg production cycle (Table 2.4). The maturity stage of fish was assessed by the method of Walsh *et al.* (1990) and in most cases the stage was checked with a histological examination. Fish were caught by trawling on the Cirolana, Walther Herwig and Kings Cross and by hand lines on the Scotia and Resolution.

During early April part of the mature female population was in spawning condition (20%) but this figure rose rapidly so that by the middle of the month 64% were spawning. In the first two collections no fish were in spent condition but by early May 4% appeared spent. However, by early June the proportion of spent fish increased to more than 30% of the population.

In conclusion the historical data and the most recent observations from the triennial survey are consistent with a two month spawning duration. A more rigourous estimate of D_S, using the method described by Rijnsdorp (1989), could be made with larger samples collected over the temporal and spatial range of the egg production survey area. However, the resulting estimate of D_S is unlikely to be less than one month or more than three months which would imply a correction for atresia of +/- 3.7% around the 1992 estimate of 7.5%.

HORSE MACKEREL

The horse mackerel spawning season in the western area is approximately 3 months, but the duration of spawning of an individual horse mackerel is regarded as much less. In the North Sea area, where the start of spawning seems to be much more synchronised, the spawning season is only about 70 days (Eltink, 1992). The duration of

spawning of an individual horse mackerel could be 70 days or may even be less.

2.8 Revision of the 1980 Egg Survey Data Point

Nichols (WD 1994) re-examined the evidence for rejection of the third survey data point for mackerel. The evidence included that presented by Darby (Appendix 1 Anon., 1993b) to the assessment working group. This shows that the lower SSB for 1980 based on accepting the rejected egg survey point is in better agreement with the historical VPA than the egg survey SSB which was used.

It was concluded from the re-examination that one of the strongest arguments used to reject the 1980 third survey data point was that the bimodal spawning curve was not expected and did not conform to the normal features of egg production curves for other species. Since then, in 1989 and in 1992 there has been some evidence that a bimodal egg production curve may be a feature in some years in the Western mackerel. The workshop were unable to form a view on whether or not to recommend reinstatement of the third survey data point. Instead it was recommended that the SSB, based on the egg production curve which includes that point, should be calculated and provided for the assessment working group meeting in June 1994.

2.9 Sampler Calibration

The high speed samplers used on these surveys are all the Gulf III type described by Gehringer (1952). However deficiencies in the original design have been recognised and numerous modifications have been made over the past forty years. The changes have tended to be made at a national level and in an *ad hoc* fashion resulting in some differences which may affect sampling performance. For example the shape and angle of the nose cone, the length and thus area of the filtering net and whether the sampler is enclosed or naked, are all features likely to affect performance. Similarly there are differences in the design and positioning of either the electronic or mechanical recording flow meter used to measure volume accepted. These flow meters are normally calibrated in either a flume or towing tank to give revolutions per metre. Unfortunately when such flow meters are mounted in the mouth opening of the sampler, the performance of the flow meter changes. This is because the shape of the nose cone generates flow profile changes along the axis of the entrance as well as radially. As a net fills with plankton so the flow rate drops and the axial and radial velocity profiles alter. As a consequence flow meters must be calibrated *in situ* in their sampler over a range of speeds and simulated clogging conditions. This was recognised at the last Plankton Sampler Workshop (Anon., 1993c). They also accepted the need to resolve the current uncertainties regarding the measurement of volume filtered in all the Gulf III type samplers in use on ICES co-ordinated plankton surveys. These uncertainties have been generated not only by the failure to calibrate flow meters *in situ*, but also by the way that they are most frequently used to calculate volume accepted. Generally their performance in free flow, in the sampler at sea has been measured and compared against their performance on a sampling station. This is then used to calculate an efficiency factor for each station or as a direct measure of distance travelled. Both approaches demand either an assumption about, or a measurement of the inherent efficiency of the sampler. At its simplest the assumption is made that the sampler accepts all the water offered to it in free flow. The differing Gulf III designs have all been subjected to attempts to measure the inherent acceptance characteristics of their mouth openings. This has led to a considerable difference in the measurement of efficiency ranging from 87.8 to 130% (Milligan and Riches, 1983; Wood and Nichols, 1983; Corten, 1990; Schnack, 1992; Brander *et al.*, 1993; Nichols, pers. comm.). These differences have been generated by the calibration method, differences in experimental design and the primary calibration device used as well as the differences in the sampler design.

Some of these issues have already been addressed and partially resolved. For example it is now recognised that because of variability in axial flow profiles the pitot-static tube, in its present configuration is not an acceptable method of measuring velocity profiles across the mouth of a sampler (Schnack, 1992). Similarly if a mini-flow meter is used for primary calibration, then great care must be taken to transect this in the correct radial plane. Results from earlier wind tunnel calibrations and calibration of the German HAI sampler in the Netherlands, which suggested efficiency values of more than 120%, were regarded as questionable. Nevertheless it is recognised that an unacceptable bias still exists in the measurement of volume filtered by Gulf III samplers which appears to be in the order of ± 10%. On plankton surveys used to estimate daily egg production this error transfers directly to an error in the estimation of spawning stock biomass. For the western mackerel stock this is ± 293,000 tonnes and ± 232,000 tonnes for the western horse mackerel., based on the 1992 survey results. The Plankton Sampler Workshop proposed to resolve the problem by re-calibrating all the existing Gulf III samplers with their flow meters *in situ* using a mini-flow meter transected across the mouth opening, in a towing tank in Hamburg. This will be done March 1994. The results will be communicated to the mackerel and horse mackerel egg workshop with a recommendation on how they should be used to correct historic data sets.

For the future the Plankton Sampler Workshop has put forward a proposal to investigate alternative methods of measuring volume filtered by high speed samplers. They accept the need, both for calibration and field use, of a non-intrusive or less intrusive system than bladed flow meters.

At an *ad hoc* meeting in Lowestoft in November 1993, representatives of Valeport Ltd, Spartel Ltd and the Robert Gordon University, Aberdeen put forward their suggestions for the development of such systems. As a result a proposal has been submitted to the EC under the AIR project for a 'Concerted Action' funding of an investigation into the feasibility of using either electromagnetic acoustic or Laser/Doppler systems for flow measurement. The time scale for development of a

new system is targeted at the next triennial mackerel/horse mackerel egg survey in 1995. The proposal also includes a thorough investigation into the performance and practicability of using 'Bongo' type samplers in preference to Gulf III's on all ICES co-ordinated surveys in future.

2.10 Publication of the Results of the 1992 AEPM and DEPM

During April 1994 a report will be submitted to the EC DGXIV Fisheries Directorate describing the results of the 1992 Project No. MA 2 436 "Spawning biology distribution and abundance of the mackerel, *Scomber scombrus* and horse mackerel, *Trachurus trachurus* in the North East Atlantic".

It was decided that the results of this study and the previous 1989 comparison should be published in the ICES Cooperative Research Report Series or similar format to make the full information available to all concerned.

3 STATISTICAL ASPECTS

3.1 Sampling Strategy for AEPM and DEPM

The failure of one of the surveys in 1992 to span the full extent of high egg densities, particularly for mackerel but also to a lesser extent for horse mackerel, indicates the need for a revision of the sampling strategy. Inadequate sampling cover leads to underestimation of biomass for both AEPM and DEPM (Anon., 1993a). In the case of AEPM, bias will occur if the spatial cover is inadequate in any one of the surveys. On average, the degree of bias is of the same magnitude as for the DEPM method, but because the latter method uses data from just one survey, bias will not occur if that particular survey provided adequate cover, but may be substantial if the survey was inadequate (as in 1992). The Workshop recommends that an improved adaptive sampling scheme should be adopted in future surveys, to guard against the possibility of very atypical spawning distributions.

In addition to the problem of not covering the full extent of the atypical spawning distribution encountered during 1992, other problems have occurred in previous surveys, and should be tackled in 1995. Given the set of rules governing interpolation to unsampled rectangles, it was often impossible to cover adequately the standard sampling area during periods when only one or two vessels were available. Additionally, cover over time is arguably inadequate, given the method used for integrating the daily egg production curve by the AEPM. Spatial models offer solutions to both these difficulties, and are addressed in Section 5.

Possible approaches for an adaptive sampling strategy are discussed and explored by Fryer *et al.* (WD 1994). In this working document, several flexible sampling strategies are explored to examine their effects on bias, coefficient of variation and root mean square error (RMSE), when tested on four different modelled distributions, derived using a generalized additive model (Hastie and Tibshirani, 1990). One of the distributions was based upon the rather anomalous westerly distribution encountered on survey 3 in 1992, another approximated the most commonly observed distribution historically, while the other two represented extreme cases: one a very dense, narrow distribution centred on the 200m contour, the other a very diffuse distribution around this contour.

The conclusions from this analysis and that of Borchers *et al.* (WD 1994) were as follows:

1) When sampling along transects across the 200m contour, a stopping rule based upon on board ship evaluation of egg numbers should be applied to decide when the distributional edge has been reached (Section 6.3). This would determine when to move to the next transect. By adopting this approach, considerable reduction in bias would result relative to a more rigid sampling strategy, and sampling effort could be more efficiently deployed.
2) Considerable improvements in precision and accuracy, as measured by RMSE, can be achieved by reducing the number of transects perpendicular to the 200m contour when ship time is limited. These improvements arise because the full east/west and north/south extent of the egg distribution can be surveyed, and because any surplus ship time is used to survey areas of high egg density more intensively.
3) A model-based approach is better able to cope with a flexible survey design than the standard method. It does not require replicate sampling of individual rectangles, provides better estimates of egg numbers in unsampled areas between transects, and gives more precise estimation of total egg abundance.

3.2 Standard Error Estimation from Vertical and Oblique Hauls

In the southern part of the distribution of mackerel and horse mackerel a different sampling method has been used by Spain in 1989 and 1992, the vertically hauled CalVET net. Because it filters a much lower volume of sea water, the CalVET net has a lower probability of catching eggs in comparison to the GULF III sampler. Both GULF III and CalVET net samples have been shown to yield reliable estimates of total daily egg production and respective standard errors (Borchers *et al.*, WD 1994; Bez & Motos, WD 1994; Motos & Uriarte, WD 1994).

If data from different sampling devices are included in the same analysis standardisation of effort is required. Effort in this case is volume of water sampled. When counts are standardized for effort, a common method of analysis is to assume the counts follow a Poisson distribution and to use a generalized linear model with a Poisson error distribution, a log link function, an estimated dispersion parameter (to allow for overdispersion), and an offset equal to *log*(effort). Borchers *et al.* (WD 1994) used a generalization of this method on the mackerel egg data, in which a generalized additive model was used in preference to a generalized linear model, because egg

density was found to be non-linearly related to available covariates.

3.3 Review of Techniques other than Arithmetic Averaging for Unsampled Rectangles

The analysis of the 1992 mackerel/horse mackerel egg survey used two methods to estimate eggs/m^2/day in unsampled rectangles (Anon., 1993a and section 8.9):

- arithmetic fill-in: use the arithmetic average of eggs/m^2/day in adjacent rectangles
- geometric fill-in: use the geometric average of eggs/m^2/day in adjacent rectangles.

The arithmetic fill-in permits valid variance estimation and is compatible with the rest of the estimation procedure, which is based on arithmetic means. The geometric fill-in has no theoretical basis and does not permit simple valid variance estimation, but has the merit of being conservative.

A number of other fill-ins could be considered. For example, an arithmetic fill-in could be adopted in which each adjacent sampled rectangle is given a weight of 1/8, regardless of how many rectangles there are. This would provide a more conservative fill-in, and variance estimation would still be simple.

However, it is anticipated that a model-based method (e.g. Borchers *et al.*, WD 1994) will be used to estimate daily egg production in 1995. Such a method models eggs/m^2/day as a function of covariates such as longitude, latitude, depth and distance from the 200m depth contour. Numbers of eggs/m^2/day are then estimated for unsampled rectangles from the covariate values for those rectangles. The question of which fill-in to use does not then arise.

3.4 Geostatistical Techniques

One aim of geostatistics is to take account of the spatial distribution of a regionalized variable and the geometry of the sampling scheme. It can use the spatial structure of horse mackerel daily egg production data to improve the precision of the estimate of eggs produced. Data from ICES Divisions VIIIa, b and c were analysed, being obtained from Spanish egg surveys using the vertically hauled CalVET net (Smith *et al.*, 1985). In the Bay of Biscay, where the analysis was focused, the experimental variograms indicate that the spatial variability of the horse mackerel daily egg production increases 4 or 5 times faster along the east-west line (transect) than along the direction of the shelf edge (Bez and Motos, WD 1994).

Estimation of global quantity of horse mackerel egg production and the corresponding variance, that makes explicit use of the inferred spatial structures, is proposed for both the Bay of Biscay and the Cantabrian Sea. Because of the sampling scheme used, it is proposed to use the "composition by lines and slices terms" (Matheron, 1971). It involves combining the errors made when extrapolating the sample values along the lines, and then extrapolating the line values out into the rectangular strips (slices) on either side of the transect. A variance is calculated for each component errors; their sum gives the variance of the total error. The coefficients of variation are respectively 4% for the Bay of Biscay and 5.4% in the Cantabrian Sea. The CV's are very low compared to CV's obtained by ignoring spatial correlation (15% in the Bay of Biscay). This shows the gain that can be achieved when the regionalization of the daily egg production is taken into account.

3.5 Conclusion

The working group has been made aware of rapid current developments in spatial modelling techniques in several statistics research centres in Europe. These methods have already influenced sampling design for 1995 and reduced CV estimates in the 1992 surveys.

The Workshop recommends that the application of this research to spawning stock biomass estimates by ichthyoplankton surveys be encouraged.

4 ANALYSIS OF THE 1992 DEPM FOR SOUTHERN HORSE MACKEREL

4.1 Division VIIIc and Sub-division IXa north (Spanish Area)

4.1.1 Revised Batch Fecundity

In the surveys carried out by Spain in Division VIIIc during 1992, ovaries with hydrated oocytes were taken and most of them were processed for histological study. For determining the batch fecundity an adaptation of the stereological method was applied (cf. Manual of the Daily Egg Production Method, 1992. Appendix A, Anon., 1993a). The batch fecundity value estimated per gramme fish weight F_{bw} was (Anon., 1993a and Porteiro *et al.*, 1993) is given below:

Area	F_{bw} (eggs/g)	SE	n
Division VIIIc	160.2	17.89	13

The value obtained is significantly different from that obtained for the western horse mackerel in 1992 (Anon., 1993a), estimated by the standard gravimetric method (Hunter *et al.*, 1985):

Area	F_{bw} (eggs/g)	SE	n
Total Western Area	209.1	5.39	162

Doubts emerged as to whether the difference in the batch fecundity values between the sea areas might be due to the methods used, the scarcity of the samples in the southern area, biological differences or other causes. These doubts have not been resolved.

To determine whether the discrepancies found in the batch fecundity estimates might be due to the use of two different methods, a set of 18 ovaries from western horse mackerel was analysed (Lucio and Pérez, WD 1994).

These ovaries had been collected by the Netherlands and processed for determining the batch fecundity, according to the standard gravimetric method (Hunter *et al.*, 1985). The samples were also processed histologically by the Netherlands (RIVO) and slides were sent to Spain (AZTI/SIO and IEO) to be analysed according to the stereological method.

The gonad volume of the 18 samples was estimated by applying the linear relationship between gonad weight (GW), and gonad volume (GV), in formalin, and applied to southern horse mackerel (Pérez and Lucio, WD 1994):

$GV = 0.09177 + (0.9479 * GW) \quad n = 99 \quad R^2 = 98.43$

The mean diameter (D) value for the hyaline oocytes in slides used in the previous stereological estimate was reexamined. From this study, the previously assumed D value of 0.9 mm did not seem to be correct. From measurements of hyaline oocytes in slides the mean D value was 0.736 mm. Using this value the batch fecundity estimates for both methods were in close agreement.

Both methods were then applied to a southern horse mackerel sample obtained in 1993. The agreement between the results from both methods was within 2% (Lucio pers. comm.).

In conclusion, the stereological method may give consistent batch fecundity values for horse mackerel.

Based on these results a new estimate of the batch fecundity for southern horse mackerel in the Spanish area was carried out, in which a corrected value for the "diameter" of the hyaline oocytes in slides was used (i.e. 0.736 instead of 0.9 mm). A new batch fecundity per gramme fish weight F_{bw} was estimated from a regression of batch size on fish weight. The regression was forced through the origin (Figure 4.1). The values of F_{bw} are presented in the following table:

Area	F_{bw} (eggs/g)	SE	n
Division VIIIc	195.9	21.88	13

This revised value for southern horse mackerel in the Spanish area is close to that obtained for the western horse mackerel in 1992 (209.1 eggs/g).

4.1.2 Revised Biomass Estimate in Spanish Area

The estimate of spawning biomass of horse mackerel in the Spanish area was revised. Total Daily Egg Production was estimated following the methodology of Pennington (1983) by blocks (ICES rectangles) as explained in Anon. (1993a). The resultant estimate was similar but the variance slightly decreased (Motos & Uriarte, WD 1994). In addition, the revised batch fecundity was also used (Lucio & Perez, WD 1994). A spawning biomass of 398,000 tonnes (CV = 0.33) was eventually estimated. This estimate reduces by 18% the previous spawning biomass estimate of 487,000 tonnes (Anon., 1993a; Porteiro *et al.*, 1993) (Table 4.1).

4.2 Sub-division IXa central-north, central-south and south (Portuguese Area)

In this area (41°50'N, 9°06'W to 36°40'N, 7°25'W) the 1992 DEPM survey was conducted on board RV "Noruega" from the 14th of February to the 20th of March to coincide with the expected maximum intensity of horse mackerel spawning. The 1992 DEPM was a secondary aim of a monitoring groundfish survey the main purpose of which was to study the juvenile and adult distribution and abundance of the main commercial species (carried out under EC FAR 1.203). On this survey fishing hauls were carried out during daylight and plankton hauls during day and night.

4.2.1 Daily Stage I Egg Production

Eighty-six plankton samples were collected from 22 east-west sampling transects, 20 n. miles apart. The sampling stations were placed 5' and 10' apart from each other (Figure 4.2) in order to fit with the bottom topography, as explained in Farinha and Borges (WD 1994).

Following Smith and Richardson (1977), the plankton was collected using a 60 cm aperture Bongo net, with 335µm mesh, by oblique hauls down to 200 meters or according to the depth of the sea bed.

Calibrated flowmeter readings were used to estimate the volume filtered per haul, to raise the number of eggs per haul to the number per m^2. On shore the eggs and larvae were separated from the zooplankton sample. The identification of stage I horse mackerel eggs was difficult for the Portuguese planktologists. For this reason the samples were exchanged with a Spanish colleague who confirmed that stage I eggs were present in the samples. However, it was noted that the eggs were in bad condition with the oil droplet divided into many small globules probably due a fixation shock (Solá, pers. comm.). It was decided to circulate the samples in order to make the results available as soon as possible (Anon., 1993a). During 1993 a Dutch colleague examined the samples and confirmed the bad quality of the eggs and the dispersion of the oil droplets. This seems to have been due to the treatment given onboard the ship (Vingerhoed pers. comm.). To fix the samples, the cod-end contents, with seawater, were immediately poured into new jars. The sample was fixed by adding neutralized 40% formaldehyde solution to make a final solution of 4% formaldehyde. Back at the laboratory on shore, the samples were filtered from the previous solution and put in a new solution of 1.1 litre of 40% formaldehyde and 10 litre of distilled water, neutralized with borax.

This workshop recommends that in future the fixation and preservation of the plankton sample should follow the manual (see Section 8.7).

The results indicated that the highest egg numbers were collected on the shelf-edge (depth 200 meters or more), similar to the horse mackerel in the Bay of Biscay and South- and West of Ireland (Figure 4.3).

The area covered was divided into two strata, one positive and another negative since the stations sampled closer to

the coast were negative for horse mackerel eggs and the off-shore stations positive. Only the hauls from the positive stratum were used in the calculations.

The observed number of stage I eggs in each station was raised to numbers per m^2 using the calibrated flowmeter readings and sampled depth. The age of the eggs was estimated using the development equations for horse mackerel, given by Pipe and Walker (1987). The number of eggs produced per m^2 per day has been calculated by multiplying by 24 hours and dividing by the age of the eggs in each station. The estimated number of stage I eggs per m^2 per day in each station was averaged by ICES half rectangle and the mean raised to the area of the half rectangle as described in Anon. (1993a).

The stations situated exactly on degrees of latitude or half degrees, overlapping the separation of rectangles were attributed to the rectangle to the north and to the west, which ever was the case. The total daily egg production was obtained by summing the half rectangles results.

The total variance was the sum of the variance in each ICES rectangle, which was calculated using the standard method version I, described in Anon., 1993a. The hauls with zero observation in the positive stratum were included in the analysis by the addition of 0.1 to all values, as recommended.

In the Portuguese area a total daily egg production of 2.42×10^{12} stage I eggs (SE = 6.19×10^{12}, CV = 10.73) was estimated using the standard analysis. Nevertheless the presence of zero hauls and high positive hauls in the same rectangle is frequent which indicates that it is not appropriate to group the hauls by ICES half rectangle. Another statistical approach should be used for the 1995 data from this area to estimate the total daily egg production and its variance (see Section 8.11).

4.2.2 Revised Spawning Fraction of Females

The estimation of the spawning fraction of females in this area was based on 10 bottom trawl hauls carried out during the same survey, only during daylight. A total of 405 histological slides of the same number of ovaries has been examined. The presence or absence of the four main oocyte maturity stages, associated with spawning, was recorded and presented at the previous Workshop meeting (Anon., 1993a). The results indicated a high percentage presence of the migratory nucleus stage (19.3%) compared with the other areas. The reason for that was that the methodology of attributing this stage was different from the one recommended by the Workshop. The slides were then exchanged with another colleague (RIVO) which resulted in a 4.0% spawning fraction of females, which is a much lower value than estimated previously (Borges *et al.*, 1993). Nevertheless doubts are expressed in Section 2.3 about the appropriateness of the use of the migratory nucleus stage to define the spawning fraction in horse mackerel.

4.2.3 Biomass Estimate in Portuguese Area

The batch fecundity estimated (Borges *et al.*, 1993) was of 170.0 eggs per gramme female for the area, with a standard error of 8.9 eggs per gramme. According to the results in the previous sections a preliminary estimate of the spawning biomass in the Portuguese area may be obtained using the total daily egg production estimated in the area and the spawning fraction obtained in the Spanish area, assuming the sex-ratio to be 0.5. This approach gives a horse mackerel spawning biomass of 360,000 tonnes. Nevertheless the precision of this estimate is very low and it should be revised in a future statistical analysis.

4.3 Divisions VIIIc and IXa

4.3.1 Biomass Estimate of Southern Horse Mackerel

At present no spawning biomass estimate can be calulated for this area, for several reasons which include the interval of 3 weeks between the surveys covering the VIIIc and IXa, and the need of further statistical analysis of the data in both areas.

5 EVALUATION OF THE 1992 EGG SURVEYS (AEPM and DEPM)

In 1992 the EC DGXIV sponsored a Project No. MA 2 436 "Spawning biology, distribution and abundance of the mackerel, *Scomber scombrus* and horse mackerel, *Trachurus trachurus* in the North East Atlantic". Funds from this project particularly enabled the University of Aberdeen to charter a commercial pelagic trawler MFV "Kings Cross" which undertook a large part of an adult fish survey at the peak of spawning that is necessary for application of the DEPM. Support was also provided to the following laboratories for additional costs related to the 1992 surveys and studies on fish spawning biology:

SOAFD Marine Laboratory, Aberdeen, UK Scotland.
Fisheries Research Centre, Dublin, Ireland.
RIVO IJmuiden, Netherlands.
IFREMER, Centre de Nantes. France.
AZTI-SIO, Sukarrieta, Spain.

Surveys undertaken by IEO, Spain and IFMK, Germany and MAFF, England and Wales also contributed to this study. Resources were also made available for a study on new methods of statistical analysis carried out by the Scottish Agricultural Statistics Service, UK Scotland.

5.1 Integration of the Annual Egg Production Curve

A crucial part of the Annual Egg Production Method (AEPM) is the integration of the daily egg production curve to estimate total egg production. Essentially, the integration process is as follows:

- Daily production is estimated for 4 or 5 survey periods throughout the spawning season.
- Daily production outside the survey periods is estimated by linear interpolation / extrapolation.

- Total production is estimated by summing the daily production estimates over the spawning season.

To date, critical assessments of the AEPM have tended to concentrate on the estimation of daily production within each survey period and to ignore the integration of the daily production curve. Two aspects of the integration process were investigated by Fryer and Ross (WD 1994):

- Its robustness to the inclusion / exclusion of a particular daily production estimate.
- The potential bias caused by estimating the production curve using only 4 or 5 daily production estimates.

In the analysis of the 1992 mackerel egg survey, there was some debate on how to treat the German and Scottish surveys in April / May (Anon., 1993a; Walsh, WD 1993). The surveys ran from 13 April - 30 April and 20 April - 5 May respectively. Although the surveys overlapped, the daily production estimate for the German survey was almost twice that of the Scottish survey, possibly indicating an early peak in spawning. Eventually, the two surveys were combined (Anon., 1993a).

The effect on the total production estimate of treating the German and Scottish surveys in 4 different ways was assessed: 1) the German and Scottish surveys were combined, as in Anon. (1993a); 2) the German and Scottish surveys treated separately, and assumed to run from 13 April -noon 25 April and noon 25 April - 5 May respectively; 3) the German survey was omitted; and 4) the Scottish survey was omitted.

The total production estimates and the percentage differences from the estimate with the German and Scottish surveys combined were:

	Estimate	%Difference
G + S combined	1.94	0.0
G + S separate	1.80	-7.2
G omitted	1.45	-25.3
S omitted	1.98	2.1

Omitting the Scottish survey has little effect on the total production estimate, whereas omitting the German survey reduces the total production estimate by 25%. In fact, omitting the German survey means that the AEPM and DEPM estimates of total stock biomass are comparable in both 1989 and 1992.

Six simple but plausible daily production curves were defined, and four sampling strategies, modelled on the 1992 survey, 'implemented'. The resulting bias in the AEPM due to integrating the daily production curve varied between -8.0% and +1.7% if the curve was assumed to have a single peak. If a second peak was allowed, bias varied between -26.2% and +11.6%. This gives an indication of the possible range of bias in the method in the absence of sampling variation. The work assumes that daily egg production is estimated without bias in each of the survey periods; further bias can be anticipated for example because geographic coverage is not perfect, because of spatio-temporal interactions (e.g. one survey might move north as the centre of egg production moves north), and because the endpoints of the spawning period may have been poorly estimated.

It is important that as many time periods are sampled as possible. Of course, a danger in increasing temporal coverage is that spatial coverage might be reduced accordingly. A failure to cover the entire spawning area within a survey period leads to a different type of bias.

Each survey contains information on how daily production changes with time within a survey period. In principle, such information might be used to fit a spatio-temporal model to the daily egg production data, for example using generalized additive models (Hastie and Tibshirani, 1990), which were used successfully by Borchers *et al.* (WD 1994) for the DEPM. This might reduce bias due to interpolation or extrapolation, in both space and time. Good spatial and temporal coverage will still be required, as temporal and spatial effects are often confounded and there are likely to be temporal/spatial interactions.

Although the above work concentrated primarily on the problems with the 1992 mackerel survey, the conclusions apply equally to the horse mackerel survey.

5.2 A Spatial Model for Egg Density and its Application to the DEPM

Estimates of the egg density component of the DEPM have contributed over 50% of the variance in the biomass estimates for both mackerel and horse mackerel. Spatial modelling allows precision of the egg density estimate to be improved. One approach using generalized additive models (GAMs) was investigated by Borchers *et al.* (WD 1994). Earlier, Pope and Woolner (1985) had fitted quadratic response surfaces to latitude, longitude and time, and Borchers *et al.* (WD 1993) considered a generalized linear model of density as a function of depth. The usefulness of such approaches arises from the fact that some of the variation in egg density is due to the variation in covariates (such as time, position and oceanic conditions) to which egg densities are closely related.

GAMs provide a particularly powerful means for modelling variation in egg density as a function of the covariates because these models accommodate functional forms of any shape, and to a large degree allow the data to determine the most suitable shape to use.

For modelling mackerel egg numbers, a log link and an overdispersed Poisson distribution with dispersion parameter 9.8 was found to be adequate. This distribution is consistent with the case that eggs are in clusters rather than randomly dispersed locally. The model incorporated non-linear effects of latitude, longitude, distance from the 200m contour and bottom depth, and linear interactions between latitude and longitude and between latitude and bottom depth.

Horse mackerel egg data proved more difficult to model. It was necessary first to model presence/absence of eggs, and second to model egg density given presence, in the spirit of Pennington's (1983) method. It was also necessary to model the Spanish non-zero egg count data separately from the non-Spanish data. The presence/absence data were modelled as a non-linear function of latitude and sea surface temperature, and as a

linear function of longitude, bottom depth and gradient. Given presence, the number of eggs for the Spanish data were modelled as a non-linear function of latitude, bottom depth and gradient, and as a linear function of longitude and time of day. The non-Spanish non-zero counts were modelled as a non-linear function of longitude and time of day and as a linear function of latitude, bottom depth and date.

Resulting egg density estimates, with standard errors obtained by bootstrapping, are given in Table 5.1 (mackerel) and Table 5.2 (horse mackerel). Also given are the corresponding estimates of stock biomass. In Table 5.3 the various estimates of stock biomass in 1992 are summarized.

For mackerel, using a GAM in place of the stratified approach reduced the CV of the egg abundance estimate from 7% to 4%. The corresponding decrease in the CV on the biomass estimate was from 13% to 9%. The revised variance of the egg abundance estimate accounts for just 25% of the variance of the biomass estimate instead of almost 60% previously.

For horse mackerel, use of a GAM reduced the CV of the egg abundance estimate from 18% to 9%, with a consequent decrease in the CV of the biomass from 22% to 18%. The variance of the egg abundance estimate contributes 33% of the variance of the biomass estimate, compared with almost 70% previously.

The advantages of using GAMs are as follows:

1) The method is very flexible. It does not assume linear relationships between the predictors and the modelled variable, and there is a choice of link functions and of assumed error distribution. As an example of the latter, it was found that a model that assumes a constant coefficient of variation in egg numbers is not optimal; past work has assumed that the CV is constant.
2) Substantial improvements in precision were achieved using GAMs. There may be scope for reduction in survey effort and therefore costs, if historic levels of precision are acceptable.
3) Interpolation (and to a very limited extent, extrapolation) can be carried out simply and reliably, thus avoiding the issue of how to fill-in when some grid squares were not sampled (e.g. whether to use geometric or arithmetic means).
4) The method has the potential to allow change over time as well as variation over space to be modelled. This could yield substantial improvements if applied to the annual egg production method.
5) The method does not require a random survey design, so that variation from the intended design is not problematic, provided there is cover at some level throughout the spawning area. The method can also use data from additional effort targeted in areas of high egg density to reduce the CV's.

The disadvantages of using GAMs are:

1) More sophisticated software and greater statistical expertise are required than for the methods used to date.
2) Bootstrapping of GAMs, to quantify precision of estimates, is very computer intensive.

These disadvantages incur a cost that is small related to the cost of achieving a similar increase in precision through increased survey effort.

5.3 Comparison of the Biomass Estimates for the AEPM and DEPM

Table 5.3 shows that the estimated biomass obtained from the GAM approach is 14% lower for mackerel and 6% higher for horse mackerel than obtained previously for the DEPM. Thus the new mackerel estimate differs even more from the AEPM method. The new horse mackerel estimate is in good agreement with those from the AEPM. The CV's for the GAM estimates are slightly lower than those on the AEPM, but it should be noted that the AEPM as implemented does not incorporate the uncertainty involved in integrating the egg production curve. Implementation of a spatio-temporal GAM for the AEPM would allow this uncertainty to be quantified, and will inflate the CV. To offset this, spatial modelling of the egg data will improve precision. It is unclear at this stage whether AEPM or DEPM would offer the greater precision, given the respective levels of effort used in 1992. However, for fixed funding to implement one method or the other, it seems clear that greater precision is attainable using the DEPM. Precision in the DEPM might be improved further by implementing spatial models for spawning fraction, fecundity and possibly female fraction.

In 1992, the survey failed to cover the full geographic distribution of eggs, especially for the mackerel, but also to a lesser extent the horse mackerel. This biased both the daily and the annual methods downwards. However, the daily method suffered from more bias because the unexpected egg distribution occurred only during the survey used by the daily method. Fryer *et al.* (WD 1994) show that the sampling strategy may be readily modified to avoid this source of bias in future surveys.

The Workshop recommends that a spatio-temporal GAM for analysing AEPM data should be developed and tested on the 1989 and 1992 data in preparation for analysing the 1995 survey data.

5.4 Choice of Methods for the 1995 Egg Production Biomass Estimates

The working group took note of ACFM's comments that "it would be premature to discontinue the AEPM until the DEPM has been shown to be successful in practice" and the opinion that "the application of the DEPM method was not successful in 1992".

Taking these two statements together the working group was obliged to apply the AEPM in 1995. Indications of resources of ship-time and laboratory analysis of samples to be made available in 1995 by various participating countries were insufficient to permit application of both the AEPM and DEPM. The working group therefore had to choose to run only the AEPM in 1995 for the western stocks of mackerel and horse mackerel. In the interests of standardisation over the whole ICES area Portugal and

Spain decided also to use the AEPM for the southern area.

5.5 Advice on the Preferred Method

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The DEPM has been applied to mackerel in 1989 and 1992. In 1989 the DEPM biomass estimate was in very close agreement with the AEPM but in 1992 was significantly lower. The Workshop believed that the DEPM accurately measured the biomass of spawning mackerel within the survey area at period 3 (survey mid-point date, 30 May 1992). Two possible components of the stock may not have been in the area at that time:

a) Fish spawning west of the spawning area (sampled by MFV Kings Cross at 52°41'N 16°05'W).
b) Spent fish that have moved out of the survey area (a possibility revealed in samples from the Norwegian Sea in June 1993).

The coefficient of variance achieved was lower than traditionally obtained for the AEPM. For the AEPM the problems of estimation of CV resulting from interpolation between sampling periods was recognised but could be resolved in future by using modelling techniques.

Assuming that full coverage of the spawning area can be achieved for estimation of egg production, the DEPM method has advantages over the AEPM on the grounds of:

(a) Reduced cost of sampling.
(b) Simpler statistical model without time as a variable.
(c) A large fishery-independent trawl sample of the adult spawning stock is obtained.

The main objection to the DEPM is that the assumed mean duration of the MNS at 24h has not been independently verified. Whilst precise timing in fish in captivity has proved elusive further studies of relative prevalence and intensity of these stages in mackerel ovaries make a duration significantly different from 24h unlikely.

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The DEPM in 1992 gave satisfactory results not significantly different from the AEPM. Statistically there is no objective means of distinguishing between the two methods.

Biologists remain concerned that in view of the low spawning fractions in horse mackerel the DEPM in this species is sensitive to errors in identification of MNS or POF oocyte stages and possible errors in assumed duration of these stages. The onset of the MNS is not as distinct in horse mackerel as in mackerel. The working group was therefore less inclined to recommend the DEPM for horse mackerel at this time.

The assumed duration of 24h for the MNS does not correspond either with the prevalence of the hyaline oocyte stage or the early POF stage. Data from the 1989 - 1992 surveys however indicate that spawning in horse mackerel is largely synchronised into a peak from 14.00h - 20.00h UTC (Motos and Muriel, WD 1994). Trawl sampling at appropriate times should therefore give a direct estimate of spawning fraction without need for further research on duration of the oocyte stages. A different adult fish sampling strategy would therefore have to be adopted when applying the DEPM for horse mackerel than for mackerel. Resource implications have not yet been assessed.

6 PLANNING OF THE WESTERN AND SOUTHERN EGG SURVEYS IN 1995

Annual Egg Production Method (AEPM)

6.1 Countries and Ships Participating

England, Germany, Ireland, Netherlands, Scotland, Portugal, Spain and Norway will participate in the mackerel/horse mackerel egg surveys in the western and southern area in 1995. Survey coverage of the western and southern areas (Figure 6.1) will be more closely interlinked than in previous years.

The survey will be split into 7 sampling periods, allowing 4 coverages of the southern area (periods 1 - 4) and 5 of the western area (periods 3 - 7). In the southern area (Figure 6.2) the annual method has not been applied before and the coverage planned represents an increase in effort compared to previous years. The widest area cover is provided during period 3 when the distribution of horse mackerel spawning is at its most widespread in the southern area. In the western area (Figure 6.3) maximum deployment of effort is during the fifth and sixth periods which coincide, respectively, with expected peak spawning of mackerel and horse mackerel in the area. 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 6.4 - 6.10.

6.2 Sampling Area

WESTERN AREA

Once again it was decided that the spatial and temporal distribution of sampling would be targeted at an adequate coverage of both mackerel and horse mackerel spawning and estimates of stage I egg production would be made for both species.

A full description of changes in the standard sampling area used in the past is given in Section 8.4. In 1995 a further increase in area has been made to take account of the unusually westerly type of distribution which occurred in period 3 1992. The new area is shown in Figure 6.3 with additional rectangles indicated by shading. No changes have been made to the northern or southern boundaries and only minor ones to the eastern boundary, the main changes are to the western boundary.

Distributions within each sampling period vary such that at the beginning and end of spawning the distributional area occupied is smaller than the standard area, while at

peak spawning it may, in some years, extend beyond it. Under these circumstances, it will be necessary in 1995 to adopt a more flexible approach to area coverage in order to sample as near to the edges of distribution as possible as well as to optimise the sampling within it (see Section 6.3). In order to aid planning of individual surveys, core areas likely to require sampling in each survey period (for both species combined) are shown in Figures 6.11 - 6.15. These are based on the historical egg distribution charts produced for the previous egg survey planning group (Anon., 1991) updated with 1992 data. In these charts rectangles containing high or medium abundances of stage I mackerel or horse mackerel eggs in any previous survey are shown to identify the likely distributional centre in 1995. The survey boundaries correspond to the areas where zero or very low egg numbers have been found in previous surveys. In the case of the rectangles which contained high egg numbers (>100 stage I eggs/m^2/day) at the outer boundary of the 1992 standard survey area, the latter has been extended by two sampling rectangles, while where medium numbers (50 - 99 stage I eggs/m^2/day) were found the area has been extended by a single rectangle.

SOUTHERN AREA

Spanish and Portuguese participants defined a standard area shown in Figure 6.2 within the coasts of Spain and Portugal for sampling the southern stock of horse mackerel and mackerel during the Annual Egg Production Method survey to be carried out in 1995. The southern and northern boundaries are limited by the 36°N and 45°N latitudes, while the western boundary by the 11°W longitude.

The standard half ICES rectangle is changed along the Cantabrian coast and the southernmost coasts of Portugal and Spain, to a quarter degree latitude by one degree longitude, because transects in those regions will be done near perpendicular to the 200 m depth contour line.

At least one haul located at the centre of each rectangle will be done, but more intensive sampling will be carried out in areas where high egg horse mackerel egg abundances are expected, based on information from previous surveys (Solá *et al.*, WD 1994).

6.3 Sampling Strategy, Gear, Procedures and Data Analysis

Most aspects under this heading have been dealt with in Section 8, however two need to be dealt with in more detail, namely - survey strategy and data analysis, since these differ substantially from previous years.

As for the southern area, the sampling gear for plankton hauls will either be a Gulf III sampler or a Bongo 20 cm mouth opening, following the procedures described in Section 8.

From analyses of 1992 egg survey data by Borchers *et al.* (WD 1994) and Fryer *et al.* (WD 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. Results from simulations on the 1992 data (Section 3.1) indicate 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 by Fryer *et al.* indicates that two important factors need to be considered when planning survey strategy. Firstly, a set of rules needs to be established for when to stop sampling a given transect so that the full distributional span is sampled while no effort is wasted outside the area of spawning. Secondly, some guide-lines need to be provided to cruise leaders on the number and spacing of transects to omit in order to best match available effort to the size of the area to be surveyed.

As a first guide to planning the distribution of sampling effort in the western area, historic egg distribution data are provided in Figures 6.11 - 6.15. The core distributional area identified for each of the different time coverages 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. 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. This represents a departure from previous survey procedure.

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 even where total coverage is expected a first pass over the area be made on alternate transects, picking up the intervening transects on the return leg.** In this way weather problems, equipment failures etc 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.

Initial investigations of modelling techniques described earlier indicate that they can give better precision with reduced sampling requirements than the previous method of working up the data. However, further work will be required in this area to develop an appropriate model which also takes account of temporal effects. This will need to be in place by the time the results of the 1995 survey become available.

6.4 Total Fecundity and Atresia Estimation

6.4.1 Total Fecundity Estimation

MACKEREL

Western area

Samples will be collected in the first three weeks of March 1995 during the MAFF western Channel groundfish survey. Sample jars filled to a standard weight with either 0.1M phosphate buffered 4% formalehyde 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 from 27 cm and above. This will correspond to about 10 fish per cm. Only fish in late pre-spawning stage 3 should be selected (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 lobe should be placed in buffered formaldehyde and the other in Gilson's fluid. Length and weight of each fish and gonad weight should be recorded and the otoliths taken.

Southern area

Spain and Portugal collect in January and February a total of 150 mackerel, covering the length range from 27 cm and above. This will correspond to about 10 fish per cm. Only fish in late pre-spawning stage 3 should be selected (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. The ovaries should be placed in buffered 4% formaldehyde. Length and weight of each fish and gonad weight should be recorded and the otoliths taken.

HORSE MACKEREL

Samples for total fecundity studies should be collected by Portugal in February and March, by Spain in March and by Germany in April. Spain and Portugal have to coordinate their sampling to ensure that about 10 ovaries for each cm group are collected from horse mackerel in late pre- spawning stage 3. Germany should collect in the western area about 10 ovaries for each cm group from horse mackerel in late pre- spawning stage 3.

The ovaries should be carefully dissected out of the fish. The ovary membrane should be pierced to allow penetration of the fixative to the lumen. The ovaries should be placed in buffered 4% formaldehyde. Length and weight of each fish and gonad weight should be recorded and the otoliths taken. The fecundity study will be carried out by both IPIMAR, Lisbon and IEO, Vigo (ovaries from the southern area) and by RIVO, IJmuiden (ovaries from the western area).

6.4.2 Atresia Estimation

MACKEREL

Western and southern area

For estimation of prevalence and relative intensity of atresia mackerel ovaries from a minimum of 90 mature fish should be collected from each survey period as given in Table 6.2.

Either a midwater trawl (peak spawning) fished close to the surface in the dark before midnight or a GOV trawl (April) should be chosen to sample the population in preference to rod and line sampling. The first 30 random selected mature females should be taken from 3 locations, spaced along the south-north axis of the egg production survey, close to the shelf edge 200 metre contour. Ovaries should be fixed in a minimum of 2 volumes of 4% buffered formaldehyde for later histological analysis. Analysis of a minimum of 50 fish per period will be divided equally between SOAFD and MAFF Lowestoft for the western area as on previous surveys (Table 6.2). Analysis of a minimum of 50 fish per period for the southern area will be divided equally between IEO, Vigo and IPIMAR, Lisbon (Table 6.2).

HORSE MACKEREL

Ovaries should be collected during all survey periods for the estimation of prevalence and relative intensity of atresia as given in Table 6.2.

Ovaries should preferably be obtained from fish caught by trawl. The first 30 random selected mature females should be taken from locations spaced along the area of high egg production. Ovaries should be fixed in a minimum of 2 volumes 4% buffered formaldehyde for later histological analysis by RIVO, IJmuiden (ovaries from the western area) and by IEO, Vigo and IPIMAR, Lisbon (ovaries from the southern area).

6.5 Coordination, Communication, Deadlines and Reporting

The coordinator of the 1995 western egg survey will be A. Eltink from the Netherlands Institute for Fishery Investigations, IJmuiden, Netherlands.

The coordinator of the 1995 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 coordinate strategies and areas of overlap if any. They should also establish a common time and radio frequency for maintaining regular contact during the cruise (2431

kHz at 18.00 UTC was found suitable for several vessels in 1992). Ships telephone, telex and fax numbers should also be exchanged between cruise leaders. 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 blank charts showing the new standard survey area will be despatched to all participants before the 1995 survey.

The coordinator of the western egg survey data base will be M. Walsh from the Marine Laboratory, Aberdeen, UK.

The coordinator of the southern egg survey data base will be A. Solá from the Instituto Español de Oceanografía, Madrid, Spain. M. Walsh 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 1995 is the dead-line for sending the egg survey results of both mackerel and horse mackerel to M. Walsh and A. Solá.

Preliminary results of total stage I egg production of both mackerel and horse mackerel together with the corresponding spawning stock biomasses will be made available to the ACFM meeting in November 1995 (if they require it).

The ICES Mackerel / Horse Mackerel Egg Production Workshop is proposed to be held 25 - 29 March 1996 at the Marine Laboratory in Aberdeen, Scotland.

Since the egg surveys of both 1995 and 1996 coincide with the usual timing of the assessment Working Group, the Egg Production Workshop recommends that the assessment Working Group should be postponed to a later date in 1995 and 1996.

7 PLANNING OF THE NORTH SEA EGG SURVEYS IN 1996

Annual Egg Production Method (AEPM)

7.1 Countries and Ships Participating

The last time the size of the spawning stock of mackerel in the North Sea was estimated based on AEPM was in 1990 (Iversen *et al.*, 1991). In Anon. (1993a) it was recommended to carry out a new egg survey in the North Sea in 1996. In 1990 the spawning stock was estimated at 78,000 tonnes (Iversen *et al.*, 1991). Single coverages of the spawning area in 1991 and 1992 indicated that the spawning stock was still at a low level (Anon., 1993a).

At present it seems that only Denmark and Norway will participate in the investigations in 1996. They will most likely allocate about three weeks each to the surveys. The exact timing of the investigations are not yet ready. However, the spawning area should be covered at least once during the peak of the spawning season which is usually during the second half of June. Usually the spawning starts mid May and ends late July.

One ship can cover the spawning area in about 12 days. A tentative schedule for covering the spawning area three times in 1996 is given below:

Country	1	2	3
Denmark	5-17 June	17-23 June	-
Norway	-	17-23 June	23 June-5 July

The period 17 - 23 June is assumed to be the peak period and will be covered by two ships each working in the area for six days. The first and third period will be covered by Denmark (RV "Dana") and a Norwegian research vessel respectively.

7.2 Sampling Area and Survey Design

Usually the main spawning area is located between 55° - 58°N and 1°- 5°E. The plankton samples will be analyzed onboard and the survey area will be adjusted accordingly. The survey grid during the coverages in 1996 might be similar to the grid applied during the second half of June 1990 (Figure 7.1). The stations close to the coast west of Denmark and Netherlands were sampled in 1990 for horse mackerel and sole eggs, and are not expected to be sampled in 1996.

7.3 Sampling and Data Analysis

The 1996 North Sea egg survey will be carried out following the standard procedure described by Iversen and Westgaard (1984) and Iversen *et al.* (1985).

The vessels will use the Gulf III type sampler or a 20 cm Bongo net. Based on the previous surveys in the North Sea there are no indications of any differences in the catch efficiency between the two samplers. Therefore the choice of gear type is not expected to have any effect on the results. A mesh size of 500 µm is used for the survey as nets with a smaller mesh size will be easily clogged.

The Danish and Norwegian vessels shall operate the samplers as in previous years, which means stepwise in depths 20m, 15m, 10m, 5m, and 0.5m. The Gulf III sampler should be towed at a speed of 5 knots and 2.5 minutes at each depth. The Bongo net should be towed at 2.5 knots for 5 minutes at each depth. The samplers shall be equipped with callibrated flowmeters.

The samples shall be placed in a standard fixative of 4% formaldehyde (see Section 8.7).

For the purpose of estimating the age of the mackerel the temperature in the surface layer at 5 m is required. It is recommended that a temperature depth profile shall be recorded at each station.

For each station information about number of stage I mackerel eggs, filtered volume and temperature at 5 m depth has to be obtained.

To obtain information on the age composition of the spawning stock of mackerel the vessels shall fish during the egg survey.

7.4 Coordination, Communication, Deadlines and Reporting

For each station information about the number of stage I mackerel eggs, filtered volume and temperature at 5m depth shall be given to the coordinator S.A. Iversen, before September 1996 and a report will be prepared for the ACFM meeting in November 1996.

8 MANUAL AEPM (including review of plankton sampling in western area)

Boxed sections represent current practice and apply to the 1995 surveys (both the western and southern area).

8.1 Target Species

The egg surveys were originally planned to cover the western mackerel and were targeted only at that species in 1977 (Lockwood *et al.*, 1981a), in 1980 (Lockwood *et al.*, 1981b) and in 1983 (Anon., 1984). Eaton (1989) showed that the surveys could also be used to estimate horse mackerel egg production. For the 1986 surveys participants were encouraged to also analyse the samples for horse mackerel eggs.

The sampling programme is now targeted at mackerel and horse mackerel and an egg production estimate is calculated for both species in both areas.

8.2 National Participation

Table 8.1 shows that participation has steadily increased since UK (England) and France began the triennial surveys in 1977 (Lockwood, 1978).

8.3 Sampling Gear

The sampler used by most participants in these surveys has been a national variant of the Gulf III type high speed sampler (Gehringer, 1952). Either the encased Dutch Gulf III, encased English 50 cm sampler or the German Nackthai have been used throughout. The only exception to this was in 1980 when paired Bongo nets were used by Germany and in 1989 (Anon., 1990) and in 1992 (Anon., 1993a) when all sampling by Spain was carried out with a vertically hauled CalVET net.

The Gulf III type samplers are towed at 5 knots, the Bongo sampler at 2-3 knots and both are deployed on a double oblique tow from the surface to sampling depth and return.

Until 1986 a 20 cm diameter mouth opening was standard on all the Gulf III type samplers used. On the 1986 survey, and subsequently the sampler used by UK (England) has been fitted with a 15 cm diameter mouth opening in accordance with an agreement to reduce sample size if required.

The standard samplers acceptable for use on these 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 filtring 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).

Calibration of the Gulf III type samplers is fully described and discussed in Section 2.9.

8.4 Standard Area

Prior to the 1977 egg surveys some information was available on the timing and distribution of mackerel spawning in the western area (Johnson, 1977). Based on that information the plankton sampling in 1977 and 1980 was designed to cover the known mackerel spawning areas from southern Biscay to west of Ireland between March and July. Following the 1977 and 1980 surveys the spatial and temporal distribution of mackerel spawning was more clearly defined. Consequently for the analysis of the 1980 survey data, geographical limits for the surveyed area were defined (Lockwood *et al.*, 1981b). These limits excluded some sampling carried out to the south-east of Ireland north of latitude 51°N. The area was also divided into rectangles of $1/2^{\circ}$ of longitude by $1/2^{\circ}$ of latitude. The standard area defined for the 1980 surveys remained the same for the 1983 surveys.

The first formal planning workshop, for the 1986 surveys, redefined the standard area (Anon., 1985). The southern limit was moved north to 45°N and the northern limit to 55°N with a recommendation that some exploratory sampling be carried out between latitudes 55°N and 56°N along the shelf edge. The western boundary, between latitudes 51°N and 54°N, was moved from longitude 15°W to 14°30'W. The eastern boundary was extended by two or three rectangles to the east across the Celtic Sea.

Planning for 1989 surveys was targeted at horse mackerel egg production as well as mackerel (Anon., 1988). As a consequence the southern boundary of the standard area was extended to 44°30'N. The eastern and western boundaries remained the same, but the northern boundary was moved to 56°N to take account of a potential northerly shift in mackerel spawning. A recommendation was made to sample opportunistically north of 56°N (Anon., 1988).

As a result of sampling north of latitude 56°N in 1989 the planning workshop for the 1992 surveys (Anon., 1991) extended the standard area to 58°N. The southern boundary was moved to 44°N with the inclusion of three rectangles between longitude 1°30'W and 3°W. The eastern boundary remained the same but because of increased mackerel spawning west of Ireland in 1989 the western boundary was returned to 15°W between latitudes 51°30'N and 54°N. The standard area used in the 1992 surveys is defined in Figure 8.6 of Anon. (1991).

The standard areas for the western and southern surveys for 1995 is defined in Section 6.2 of this report.

8.5 Sampling Strategy

The temporal coverage of the western area in each of the survey years in shown in Figure 8.1. The survey number indicates one survey which generated a point on the annual egg production curve for mackerel. However the whole western standard area was not always sampled on each survey. For this information it is necessary to refer to the relevant reports for each year. Temporal coverage has generally been dictated by the availability of ships time, which has fluctuated over the years, although some positive decisions to change have been taken. Plankton sampling during March in the western area has been discontinued since 1986 because it is not necessary in order to determine the annual production of either mackerel or horse mackerel eggs. Similarly extending sampling beyond the middle of July is no longer considered necessary.

The current sampling strategy has evolved over the past sixteen years as a result of experience, increased knowledge and changes in survey requirements and availability of resources. It will be useful for the future to summarise that evolution from the deliberations of the various workshops and planning reports.

In 1977 very limited resources were available with only two countries participating. With a large area to cover over a long time period plankton sampling density was poor. A basic pattern of single hauls $^1/_2$° of longitude apart on rows 1° of latitude apart was adopted. A small amount of additional sampling was achieved in Biscay and on one survey in the central area.

By 1980 the standard area was divided into sampling rectangles of $^1/_2$° of latitude by $^1/_2$° of longitude. Sampling was targeted at a single station at the centre of each of those rectangles. (Sampling, west of Ireland, on the first survey of that year was more intensive).

The basic strategy of taking one sample in the centre of each of the rectangles was continued in 1983. In some areas, where mackerel eggs were more abundant, more than one sample per rectangle was taken. This was achieved either by repeated sampling by the same vessel, or on some surveys by more than one vessel sampling the same rectangle.

For the 1986 surveys a planning group adopted a formal sampling strategy, still based on rectangles but with the standard area divided into high and low sampling strata (Anon., 1985). The strata were designated on the basis of the occurrence of mackerel eggs on previous surveys. The season was divided into three survey periods and the aim was to sample at a ratio of 2 samples to 1 in favour of the potentially high egg density stratum during the middle survey. Samples would still be taken in the centre of each rectangle. The replicate sampling would be achieved with either two consecutive samples or a single coverage of the whole area and a return to take additional samples in the high stratum.

This strategy was successful in 1986 (Anon., 1987) although over sampling of the low stratum did occur. The season was eventually divided into four periods with the additional sampling occurring in periods two and three.

For the 1989 surveys a more flexible strategy was adopted without the rigid designation of high and low strata. Instead guidance was given, on the basis of previous surveys, on where the highest egg densities were likely to occur in any time period (Anon., 1988). The surveys were now targeted at both mackerel and horse mackerel therefore the spawning distribution of both species had to be considered. A 'common sense' strategy was adopted to allow those taking part in the survey the flexibility to make decisions about replicate sampling based on the guidance and shipboard enumeration of samples. There was also an instruction to reduce the number of 'zero' observations compared with previous survey years.

Sampling for the new Daily Egg Production Method, (DEPM) was being incorporated into the 1989 surveys for the first time. As a result, and with additional ships time available, the spawning season was split into five sampling periods with maximum effort being put into the three central periods. In practice significant replicate sampling was only achieved on periods 3 and 4. Furthermore because of a failure to follow sampling protocol during period 1, when samples were not taken in the centres of rectangles, this survey result, which gave an unacceptably high egg production for both species, was rejected.

Once again in 1992 sampling was targeted at both mackerel and horse mackerel. Under the terms of an EC contract plankton sampling to allow the DEPM to be applied to both species had to be incorporated. Five survey periods were planned with the maximum effort aimed at the third period which was the expected peak period for both mackerel and horse mackerel egg production. The results from this period only would be used for the DEPM (Anon., 1991).

A flexible approach was again adopted to replicate sampling within a rectangle. However there was a firm recommendation that additional sampling should be carried out in areas where high densities of either mackerel or horse mackerel eggs could be expected. As guidance to the areas where these high densities were likely to occur, charts showing the maximum contribution to egg production of either species in each time period were provided (Anon., 1991). In an attempt to improve spatial resolution, replicate samples within a rectangle would not be taken in the centre of those rectangles. Instead they were to be evenly spaced in an east-west direction. This marked a significant change in

strategy from all previous survey years. In areas where, and at times when, low egg densities could be expected, only alternate rows of rectangles would be sampled.

Because of operational problems surveys 1 and 2 in 1992 had to be combined, therefore only four coverage's were achieved. A satisfactory level of replicate sampling was achieved with this strategy, in particular during period 3 (second survey). However it was generally agreed that this was at the expense of flexibility and probably resulted in a failure to detect and thus react to high densities of mackerel eggs at the western edge of the area during the second survey.

The sampling strategy in the western and southern area in 1995 will be targeted at the AEPM only. A flexible approach will again be adopted to the area sampled and to replicate sampling. This will be based on the charts for each period being updated by the inclusion of the 1992 data (see Section 6.3). Sampling of horse mackerel eggs should be spread uniformly throughout the whole 24 hour period.

8.6 Sampling Depth

This has been subjected to both formal changes and for operational reasons to *ad hoc* changes over the period of these surveys.

In 1977 the maximum sampling depth was 100 m or to within 2 m of the bottom where the bottom depth was less than 100 m.

In 1980 the maximum sampling depth was changed to 200 m. However paired Bongo nets used on the first survey were deployed to a maximum depth of 150 m only. The change to 200 m was based on the observation of Coombs *et al.* (1981) which showed that, before the establishment of a thermocline, the majority of mackerel eggs are found between the surface and 200 m depth.

In 1983 the maximum sampling depth was changed back to 100 m. This was the result of further vertical distribution studies which showed that the majority of mackerel eggs were found above 100 m. These studies also showed that in the presence of a 3°C thermocline mackerel eggs were all found above that thermocline. As a consequence, in the presence of a 3°C thermocline or greater, maximum sampling depth was confined to 20 m below the thermocline. As an exception to the planned strategy, on the first survey in March the sampler was deployed to a maximum depth of 200 m.

For the 1986 surveys it was recommended that in the absence of a thermocline, sampling should be to the bottom or to 200 m, whichever is the shallower. This resulted from further examination of vertical distribution data which showed that, particularly during the early season, there could be some under-sampling of mackerel eggs if sampling was confined to just 100 m depth. The condition under which sampling could be confined to 20 m below the thermocline were more clearly defined. In the presence of a thermocline, of at least 2°C over 10 m in depth, sampling should be limited to 20 m below the thermocline (Anon., 1985).

The above depth sampling strategy (Anon., 1985) remained extant for the 1989 and 1992 surveys although in 1989 France and Ireland sampled to only 150 m. As a result of some further vertical distribution studies on mackerel eggs reported in Anon. (1993a) a small change in the procedure for future surveys is recommended. That change is that 'in the presence of a thermocline greater than 2.5°C in 10 m depth (previously 2.0°C in 10 m depth) sampling can be confined to a maximum depth of 20 m below the thermocline'.

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 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 *(the instruction in Anon. (1991) to integrate over the bottom depth when the bottom is 200 m or less is wrong and should ignored)*

8.7 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 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.

8.8 Egg Identification, Staging and Ageing

The identification of mackerel and horse mackerel eggs is based on the description by Russell (1976). Exchange of samples of both species between participating countries has shown that there are no problems in identifying mackerel and horse mackerel eggs in the western area.

The eggs of mackerel should be classified into one of five morphological stages (I, II, III, IV and V) (Lockwood *et al.*, 1981a) 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, 1981a). 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.

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:

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

Further studies of mackerel egg development were carried out in relation to studies of the diel periodicity of spawning (Nichols and Warnes, 1993). Observation of the early development of the egg were more frequent in these studies and thus provide a more precise estimate of development rate then the 1977 experiments. However the observations were made over a smaller temperature range and therefore no change in the formula has been made. It is recommended however that an opportunity be sought to incubate mackerel eggs through stage I with observations at intervals more frequent than six hourly (Anon., 1993a).

The sea temperature used to calculate the duration of stage I eggs for both species has not remained the same since the egg surveys began. In the 1977, 1980 and 1983 surveys the surface temperature was used for this calculation. By 1986 many of the vessels were collecting concurrent temperature profiles during a plankton haul. As a consequence it was recommended that, when available the temperature at 20 m depth should be used for the calculation. If that was not available then the sub-surface temperature (ca. 3 m) should be used. By the 1989 survey all participants were providing the temperature at 20 m depth.

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.

8.9 Rectangle Sampling

Analysis of the 1977 survey data was not based on $1/2°$ x $1/2°$ rectangles therefore the problem of interpolating unsampled rectangles did not arise. No extrapolations to unsampled areas were made.

Since 1977 all the egg survey data has been analysed on the basis of production within each rectangle. For the 1980 surveys it was decided that interpolation would be made for unsampled rectangles within the sampled area, but that no extrapolation to unsampled areas would be made. A protocol for interpolation was established in 1980 (Lockwood *et al.*, 1981b) which has remained operative on all subsequent surveys until 1992. During analysis of the 1992 survey results it was decided to compare the result of using either geometric or arithmetic means for calculating interpolated values (Anon., 1993a). As a result of this comparison it was decided to reject the geometric mean in favour of the arithmetic mean. The historic data set has now been re-calculated accordingly. The current protocol for interpolation of unsampled rectangles is as stated by Lockwood *et al.* (1981b), but using arithmetic means of the adjacent rectangles instead of geometric means.

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.

For the 1995 surveys the preliminary calculations will follow the methods described above for the 1992 surveys. Subsequently the data will be analysed as described in Section 8.11.

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.

8.10 Sampling on the Edge of the Rectangles

The basic sampling strategy has always been either for a sample in the centre of a rectangle or two equidistant samples within a rectangle. Serious problems of analysis can arise, as in the 1989 surveys, when this protocol is not strictly followed.

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.

(This convention was not followed for sampling by Spain in 1992 when samples were allocated to the rectangle east of the line of longitude).

8.11 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) =

$$\frac{\text{Flowm-revs x Aperture}}{\text{Flowm-calib}} \text{ x Efficiency Factor}$$

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

$$\text{Eggs/m}^2 = \frac{\text{Eggs counted x Factor}}{\text{Volume filtered}} \text{ x Depth sampled}$$

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.

Numbers of eggs per m^2 are raised to numbers per m^2 per day using development equation for both species (see Section 8.8) in the following way.

For stage I **mackerel** eggs:

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

For stage I **horse mackerel** eggs:

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

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 $^1/_2°$ row of latitude using the formula:

Area (m^2) = (cos(latitude) x 30 x 1853.2) x (30 x 1853.2)

When there is more than one observation per rectangle within a sampling period, the arithmetic mean of the observed values is used.

The above procedure has applied to all surveys since the 1977 survey which was not analysed by rectangles.

The coefficient of variation (CV) of the number of mackerel eggs per m^2 per day has been estimated for each survey from 1983, following a procedure described by Pope and Woolner (1984). The estimate of CV for the 1983 survey data (1.3) derived from 68 replicated rectangles within cruises. The 1986 data had 118 such replicates but the CV remained the same. The variance from sampled rectangles is obtained by summing the squares of rectangle values and multiplying by the constant CV squared. For interpolated rectangles the procedure is the same except that the value of CV is not constant but depends on the number of rectangles used to determine the interpolated values. The same procedure was followed for the 1989 survey data when the constant CV calculated was 1.3.

The procedure changed for the 1992 survey (Anon., 1993a) based on the working document of Fryer *et al.* (WD 1993). For both species, the coefficient of variation (CV) of eggs per m^2 per day was estimated from 1992 survey by an analysis of variance of log (eggs/m^2/day) on rectangle and survey period, having excluded those rectangle/period combinations for which there were any zero hauls. These rectangle/periods were excluded to reduce the highly influential effect of zero values (Fryer *et al.*, WD 1993). This approach gave a CV of 1.1 for mackerel and 1.5 for horse mackerel. The corresponding values, based on the 1989 survey (Anon., 1990), were 1.3 for both species, but comparisons between the CV's for the two surveys are invalid due to the different treatment of zero values.

The variance of the egg production of a sampled half ICES rectangle was estimated to be:

Variance = (Area x eggs/m^2/day x CV)^2/number of hauls

where the area of the half ICES rectangle is given in m^2.

Total egg production per day was estimated by summing the production estimates for each rectangle (sampled and extrapolated) in the survey grid. Given no extrapolation, the variance of the total egg production per day is given by the sum of the variances for the individual half rectangles. Where there is arithmetic extrapolation, the variance of the total egg production per day is adjusted appropriately (Anon., 1993a).

The procedure for estimating the total annual egg production has remained essentially the same since the surveys began. The individual survey period totals are plotted against time at the mid sampling point of each survey period. Total annual egg production is then calculated by integrating the area under the resultant curve. This is equivalent to taking a weighted sum of the total daily production in each sampling period. The variance of the total annual egg production is also a weighted sum of the variances of the total daily production in each sampling period (Anon., 1993a).

The starting and finishing dates for the annual egg production curve for each species in the western area have varied for each of the survey years, because of the way they are calculated. The starting date is based on a back calculation to its spawned date of the latest stage egg of each species on the first survey. The finishing date for each species is on the last day on which stage I eggs of that species are found. Assumed starting dates have thus varied between 14 - 21 March for mackerel and 1 March - 1 April for horse mackerel. Assumed finishing dates have varied between 10 - 25 July for mackerel and 10 - 31 July for horse mackerel.

For the 1995 surveys a model based approach to data analysis is advocated. Borchers *et al.* (WD 1994) have shown the value of a spatial model using Generalised Additive Models (GAMs) in improving the precision of the DEPM. The gain for the AEPM is potentially much greater. The greatest statistical flaw in the AEPM is the method of estimation of the daily egg production curve. Observations are assumed to have been made at four or five discrete time points, and the curve is constructed by joining the observations with straight lines. Not only does this risk substantial bias (see for example Fryer and Ross, WD 1994), but the precision of this estimation process is not quantified. In reality each survey runs for many days, allowing a spatio-temporal model to be fitted to the survey data, and hence for the daily egg production curve to be estimated by GAM methods. Problems can be anticipated with this approach; for example, temporal effects will be confounded with spatial effects. However we anticipate that the reliability of the estimates will be substantially greater than for the method used in the past.

It is recommended that a Generalised Additive Model based approach, comparable to that described by Borchers *et al.* (WD 1994), is used to analyse the 1995 survey data.

To assess the gains from such an approach, and to ensure comparability with previous estimates, the 1989 and 1992 survey data should be analysed using GAMs prior to the analysis of the 1995 survey data.

It is anticipated that this analysis, and the analysis of the 1995 survey data, will require substantial commitment of a full time specialist. Without that commitment the data cannot be analysed in this way.

9 DEFICIENCIES AND RECOMMENDATIONS

1. There is an on-going problem with the identification of some stages of mackerel and horse mackerel eggs. The Workshop therefore recommends that a further egg exchange exercise for mackerel and horse mackerel will take place in 1995 by UK (England).
2. The Workshop could not decide whether the rejected 1980 egg survey data point should be reinstated. The Workshop therefore recommends that the spawning stock biomass based on the reinstated point should be calculated and be made available to the Mackerel, Sardine and Anchovy Working Group.
3. The Workshop felt that the results of the comparisons between the DEPM and the AEPM for mackerel and horse mackerel in the western area should be made more widely available than the official Report on the Contract to the EC. The Workshop therefore recommends that the data should be published as an ICES Cooperative Research Report edited by I. G. Priede and A. Eltink.
4. There is a need to improve the precision and to reduce the bias of the egg survey estimates. As a potential way forward the Workshop recommends that the application of spatial modelling techniques to spawning stock biomass estimation by ichthyoplankton survey be encouraged.
5. The Workshop decided that a spatio-temporal generalised additive model should be used for the analysis of the 1995 egg survey data. The Workshop recommends that in preparation for this analysis the method should be developed and tested on the 1989 and 1992 data.
6. Because horse mackerel spawning is synchronised there is potential bias if plankton sampling is confined to any particular time of the day or night. The Workshop therefore recommends that sampling for horse mackerel eggs should be spread uniformly throughout the 24 hour period.
7. There is a need to continue to collect the long term series of information on the batch fecundity of mackerel. The Workshop therefore recommends that hydrated oocyte ovary samples be collected for batch fecundity determination in the area 49°N - 51°N; 9°W - 11°W.
8. There was an unusual westerly distribution of spawning of mackerel during the third survey period in 1992 which was not sampled on the plankton survey because the sampling strategy was not flexible enough. The Workshop therefore recommends that for the 1995 surveys an improved adaptive sampling scheme should be adopted. This will be based on shipboard observation of the presence of stage I mackerel or horse mackerel eggs in the samples during the survey.
9. For the analysis of the 1995 egg surveys a next Mackerel / Horse Mackerel Egg Production Workshop is proposed to be held 25 - 29 March 1996 at the Marine Laboratory in Aberdeen, Scotland.
10. Since the egg surveys of both 1995 and 1996 coincide with the usual timing of the assessment Working Group, the Mackerel / Horse Mackerel Egg Production Workshop recommends that the assessment Working Group be postponed to a later date in 1995 and 1996.

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Table 2.1a Mackerel eggs by stage in total numbers counted.

	I	II	III	IV	V	Total
England	128	117	96	91	68	500
France	171	153	62	39	75	500
Scotland	146	52	186	52	65	501
Ireland	128	88	138	62	84	500
Germany	148	84	139	50	80	501
Netherlands	140	119	99	69	73	493 ***
Spain 1	147	95	129	53	76	488 ***
Spain 2	148	86	136	58	72	483 ***
Mean	144.4	99.2	123.1	59.3	74.2	
SD	12.7	28.2	34.7	14.6	5.7	
CV	8.8	28.4	28.2	24.6	7.7	

Table 2.1b Horse mackerel eggs by stage in total numbers counted.

	I	II	III	IV	Total
England	113	165	87	135	500
France	154	188	92	65	499
Scotland	129	57	236	78	500
Ireland	119	123	134	121	497
Germany	139	107	150	103	499
Netherlands	128	158	130	84	493 ***
Spain 1	119	129	140	112	486 ***
Spain 2	137	104	135	123	491 ***
Mean	129.8	128.9	138.0	102.6	
SD	12.5	38.6	42.7	23.1	
CV	9.6	30.0	30.9	22.5	

*** - Stage numbers normalised to total = 500.

Table 2.1c Coefficient of variation by sample and by egg stage of mackerel.

Sample No.	I	II	III	IV	V
2	14.5	23.4	27.9	23.2	6.9
4	12.5	49.0	22.9	36.0	6.4
6	12.7	32.9	31.4	29.3	30.3
8	0.0	63.0	31.9	33.2	15.4
10	8.3	39.6	39.4	17.2	13.3
12	20.4	14.1	41.6	45.5	14.1
14	7.4	59.6	25.6	52.8	27.4
16	11.8	16.1	38.2	35.1	13.4
18	14.7	54.0	29.4	42.4	33.3
20	19.5	28.5	43.0	22.8	10.5
Total	8.8	28.4	28.2	24.7	7.7

Table 2.1d Coefficient of variation by sample and by egg stage of horse mackerel.

Sample No.	I	II	III	IV
1	26.3	37.8	27.2	23.2
3	22.7	36.5	41.3	36.0
5	15.4	12.9	47.1	29.3
7	29.3	26.5	35.8	33.2
9	4.8	41.9	84.6	17.2
11	2.8	56.2	30.3	45.5
13	19.9	29.4	25.3	52.8
15	18.3	63.1	43.2	35.1
17	19.7	30.1	26.3	42.4
19	15.1	41.4	39.5	22.8
Total	9.6	30.0	30.9	22. 5

Table 2.2 A summary of the mean catches and the histological analysis of samples of mature spawning mackerel caught in the FOTØ trawl and by rod and line in 1992. A sequence of hauls around mid-day were made with the trawl headline on the surface, at 50m and at 100m for a period of 1h followed by a rod and line station. This sequence of hauls was repeated, centred around mid-night. Rod and line catches were raised to the average weight of fish caught on the surface.

		DAY			NIGHT		
	Headline depth (m)	Number of stations	Catch weight (kg)	± SD	Number of stations	Catch weight (kg)	± SD
Mackerel catch weight (kg)	0	4	85	80	1	237	-
	50	4	23	19	1	36	-
	100	3	8	6	1	4	-
			Proportion caught			Proportion caught	
The proportion of mackerel caught containing oocytes with migratory nuclei	Rod & Line	2	0.62	0.08			
	0	4	0.58	0.08	1	0.47	-
	50	4	0.43	0.26	1	0.33	-
	100	2	0.81	0.02	1	0.38	-
The proportion of mackerel caught with very fresh post-ovulatory follicles in the ovary	Rod & Line	2	0.42	0.08			
	0	4	0.31	0.15	1	0.07	-
	50	4	0.19	0.14	1	0.07	-
	100	2	0.07	0.07	1	0.00	-
			Number caught			Number caught	
The number of mackerel caught with very fresh post-ovulatory follicles in the ovary	Rod & Line	2	40	7			
	0	4	35	31	1	22	-
	50	4	10	12	1	3	-
	100	3	2	2	1	0	-

Table 2.3 Details of the mean weight of horse mackerel and mackerel caught in the FOTØ trawl in 1993. A sequence of three 1 hour hauls around mid-day was made with the trawl headline on the surface, at 50m and at 100m for a period of 1h. This sequence of hauls was repeated, centred around mid night the following night. Later in the cruise, the sequence of trawls was repeated around dusk at 20:00 hours GMT.

Headline depth	Day / night	Mean catch weight (kg) of horse mackerel caught (sd)		Mean catch weight (kg) of mackerel caught (sd)		Number of hauls
0	day	0	(0)	1	(1)	4
50		0	(0)	1	(1)	4
100		4	(2)	3	(2)	4
0	night	110	(11)	321	(137)	4
50		11	(11)	25	(31	4
100		1	(1)	12	(14)	4
0	dusk	9	(5)	286	(193)	2
50		4	(1)	3	(2)	2
100		13	(10)	2	(2)	2

Table 2.4 Mackerel maturity stages observed in fish caught on five research vessel cruises during the 1992 triennial survey. The maturity stages were checked by histology.

Vessel	Mid-date of collection	Latitudinal range of collection	No of hauls	% of mature females by stage			no of fish examined
				3	4-5	6	Total
Cirolana 4/92 (*)	4/4	48° - 51°N	6	80	20	0	49
Walther Herwig (*)	16/4	46° - 48°N	5	36	64	0	44
Scotia (*)	1/5	48° - 53°N	4	20	76	4	25
Kings Cross (*)	4/6	50° - 57°N	19	8	48	31	26
Resolution	4/7	49° - 56°N	13	18	71	11	92

(*) Fish with post ovulatory follicles but without hydrated oocytes were placed in the stage 4-5 group. The spent category included fish with abundant atresia and sporadic yolk vesicle stage oocytes.

Table 4.1 Daily Egg Production Method in 1992 applied on Southern Horse mackerel in Spanish area (Div. VIIIc and IXa) after revision of batch fecundity value in 1994. Pennington's method (1983) estimates (CV's are in brackets). The CV of the spawning biomass is estimated following the delta method (Seber, 1982).

Number of stage I eggs ($x10^{-12}$)	Relative Batch fecundity F_{bw} (eggs/g)	Spawning fraction S (based on migr.nucl.)	Female fraction R	Female mean weight (g)	Spawning biomass ('000 tonnes)
3.301	195.89	0.0847	0.5	193.29	397.871
(0.202)	(21.88)	(0.23)		(0.04)	(0.33)

Table 5.1 Egg density and corresponding stock biomass estimates of mackerel from the Daily Egg Production method (DEPM) obtained by fitting GAMs to the egg data. Figures in parentheses are the (standard errors) and [% coefficient of variance].

Stratum	Egg numbers (no.x10^{12})	Fecundity (eggs/g)	Spawning fraction	Biomass (tonnes x10^6)
Northern	1.049 (0.209) [19.9]	28.69 (2.74) [9.5]	0.467 (0.085) [18.1]	0.1568 (0.0448) [28.6]
Middle	7.128 (0.553) [7.8]	47.37 (2.17) [4.6]	0.573 (0.055) [9.7]	0.5370 (0.0687) [12.8]
Southern	12.333 (0.686) [5.6]	49.82 (4.36) [8.7]	0.480 (0.040) [8.4]	9.905 (1.253) [12.7]
Total	20.511 (0.905) [4.4]	1.6843* (0.1498) [8.9]		

*When this biomass estimate is multiplied by the correction factor for hyaline to average spawning biomass (0.959, which is assumed to have CV=0), the resulting biomass estimate is 1.615 x10^6 tonnes.

Table 5.2 Egg density and corresponding stock biomass estimates of horse mackerel from the Daily Egg Production method (DEPM) obtained by fitting GAMs to the egg data. Figures in parentheses are the (standard errors) and [% coefficient of variance].

Stratum	Egg numbers (no.x10^{12})	Fecundity (eggs/g)	Spawning fraction	Biomass (tonnes x10^6)
Northern	0.984 (0.108) [11.0]	212.4 (14.1) [6.8]	0.033 (0.011) [32.5]	0.2801 (0.098) [35.0]
Middle	9.461 (0.542) [5.7]	203.6 (7.5) [3.7]	0.104 (0.018) [17.5]	0.8963 (0.168) [18.8]
Southern	5.097 (1.225) [24.0]	209.5 (10.9) [5.2]	0.058 (0.016) [27.5]	0.835 (0.308) [36.9]
Total	15.542 (1.344) [8.6]	2.012* (0.364) [18.1]		

*When this biomass estimate is multiplied by the correction factor for hyaline to average spawning biomass (0.974, which is assumed to have CV=0), the resulting biomass estimate is 1.96 x10^6 tonnes.

Table 5.3 Comparison of 1992 biomass estimates (tonnes x10^6) under the AEPM and DEPM. Geom. refers to a geometric mean fill-in for unsampled squares, and Arith. to an arithmetic mean fill-in. Previous refers to the agreed estimate following the 1993 workshop (Anon., 1993a), and GAM refers to estimates obtained by generalized additive modelling of the egg data only.

			Western Mackerel			Western Horse Mackerel		
			Est.	se	CV(%)	Est.	se	CV(%)
AEPM	No atresia correction	Geom.	2.52	-	-	1.81	-	-
		Arith.	2.67	0.26	10	2.09	0.40	19
	Atresia correction	Geom.	2.76	-	-	2.01	-	-
		Arith.	2.93	0.29	10	2.32	0.45	19
DEPM	With hyaline correction	Previous	1.88	0.25	13	1.84	0.40	22
		GAM	1.62	0.14	9	1.96	0.35	18

Table 6.1 Planned research vessel deployment for the 1995 Mackerel / Horse Mackerel Egg Surveys in the western and southern area.

Coverage	Country	Area	Ship	Week number(s)	Period	Survey mid-point	Latitude to be covered
1	Portugal	South	Capricornio	3, 4	6 - 19 Feb	12 - 13 Feb	36°N-41°30'N
2	Portugal	South	Capricornio	10, 11, 12	6 Mar - 26 Mar	16 March	36°N-43°N
3	Portugal Spain Germany	South South/West West	Capricornio Cornide Walther Herwig	14 13, 14, 15 13, 14, 15	27 Mar - 16 Apr	6 April	36°N-39°N 39°N-45°N 45°N-55°N
4	England Scotland	South/West West	Cirolana Scotia	17, 18, 19 17, 18, 19	24 Apr - 14 May	4 May	43°N-49°30'N 48°30'N-56°N
5	Ireland Netherlands Spain	West West South/West	Lough Foyle Tridens Cornide	21, 22, 23 21, 22 20, 21	15 May - 11 June	28 - 29 May	49°30'N-58°N 46°30'N-50°N 43°N-47°N
6	Norway Netherlands	West West	G. O. Sars Tridens	24, 25, 26 24, 25, 26	12 June - 2 July	22 June	49°30'N-58°N 44°N-50°N
7	Scotland	West	Charter	27, 28, 29	3 July - 23 July	13 July	44°N-57°N

Table 6.2 Minimum numbers of ovaries to be sampled for **atresia** investigations during the different coverages. During each coverage 90 ovaries should be sampled in the southern area and another 90 ovaries should be sampled in the western area (for both mackerel and horse mackerel). Laboratories are indicated where ovary samples have to be sent to and where the histological analysis will be carried out. Names between brackets are the coordinators by country.

Coverage	Country	Period	Area	Mackerel		Horse mackerel	
				Minimum No. of fish	Laboratory	Minimum No. of fish	Laboratory
1	Portugal	6.2-19.2	36°-41°30'N	90	IPIMAR	90	IPIMAR
2	Portugal	6.3-26.3	36°-43°N	90	IPIMAR	90	IPIMAR
3	Germany	27.3-16.4	45°-55°N	90	SOAFD	90	RIVO
	Portugal		36°-39°N	45	IPIMAR	45	IPIMAR
	Spain		39°-45°N	45	IEO	45	IEO
4	England	24.4-14.5	43°-49°30'N	135	MAFF, IEO	180	RIVO, IEO
	Scotland		48°30'-56°N	45	SOAFD	-	IEO
5	Ireland	15.5-11.6	49°30'-58°N	30	SOAFD	-	-
	Netherlands		46°30'-50°N	30	MAFF	90	RIVO
	Spain		43°-47°N	120	IEO	90	IEO
6	Norway	12.6-2.7	49°30'-58°N	45	SOAFD	-	-
	Netherlands		44°-50°N	45	MAFF	90	RIVO
7	Scotland	3.7-23.7	44°-57°N	90	MAFF, SOAFD	90	RIVO

SOAFD in Aberdeen (M. Walsh)
MAFF in Lowestoft (P.R. Witthames)
IEO in Vigo (C. Porteiro)
RIVO in IJmuiden (A. Eltink)
IPIMAR in Lisbon (F. Borges)

Table 8.1 National participation in the western and southern egg surveys by country by year during the period 1977 - 1995.

	1977	1980	1983	1986	1989	1992	1995
France	*	*	*		*	*	
Germany		*	*	*	*	*	*
Ireland				*	*	*	*
Netherlands			*	*	*	*	*
Spain					*	*	*
UK (England)	*	*	*	*	*	*	*
UK (Scotland)		*	*	*	*	*	*
Norway							*
Spain					*	*	*
Portugal						*	*

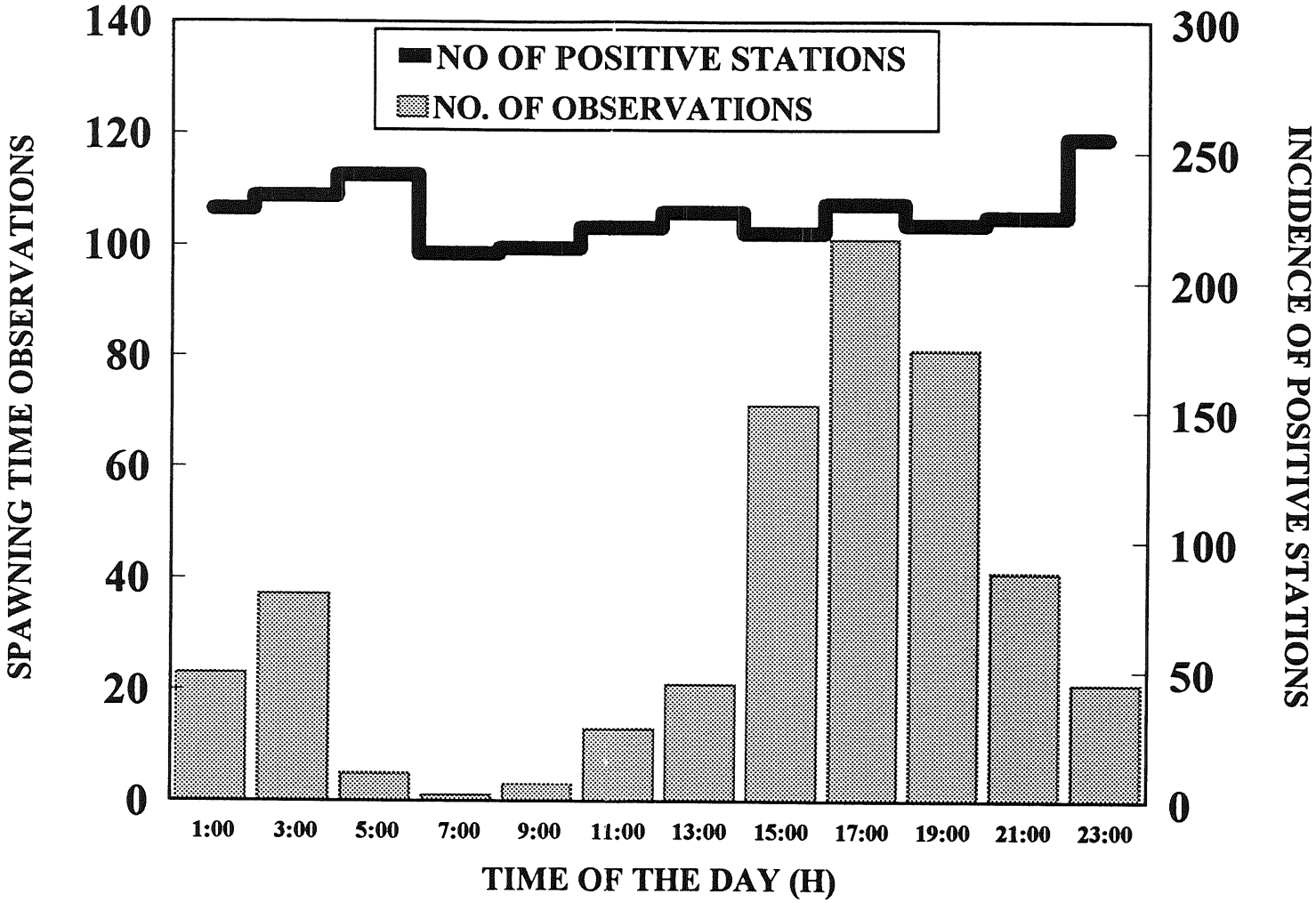


Figure 2.1 Incidence of back-calculated spawning time by two hour periods (bars). The number of samples containing horse mackerel eggs are shown by two hour periods. Based on egg data from the egg surveys carried out in spring 1989 and 1992 in the Bay of Biscay.

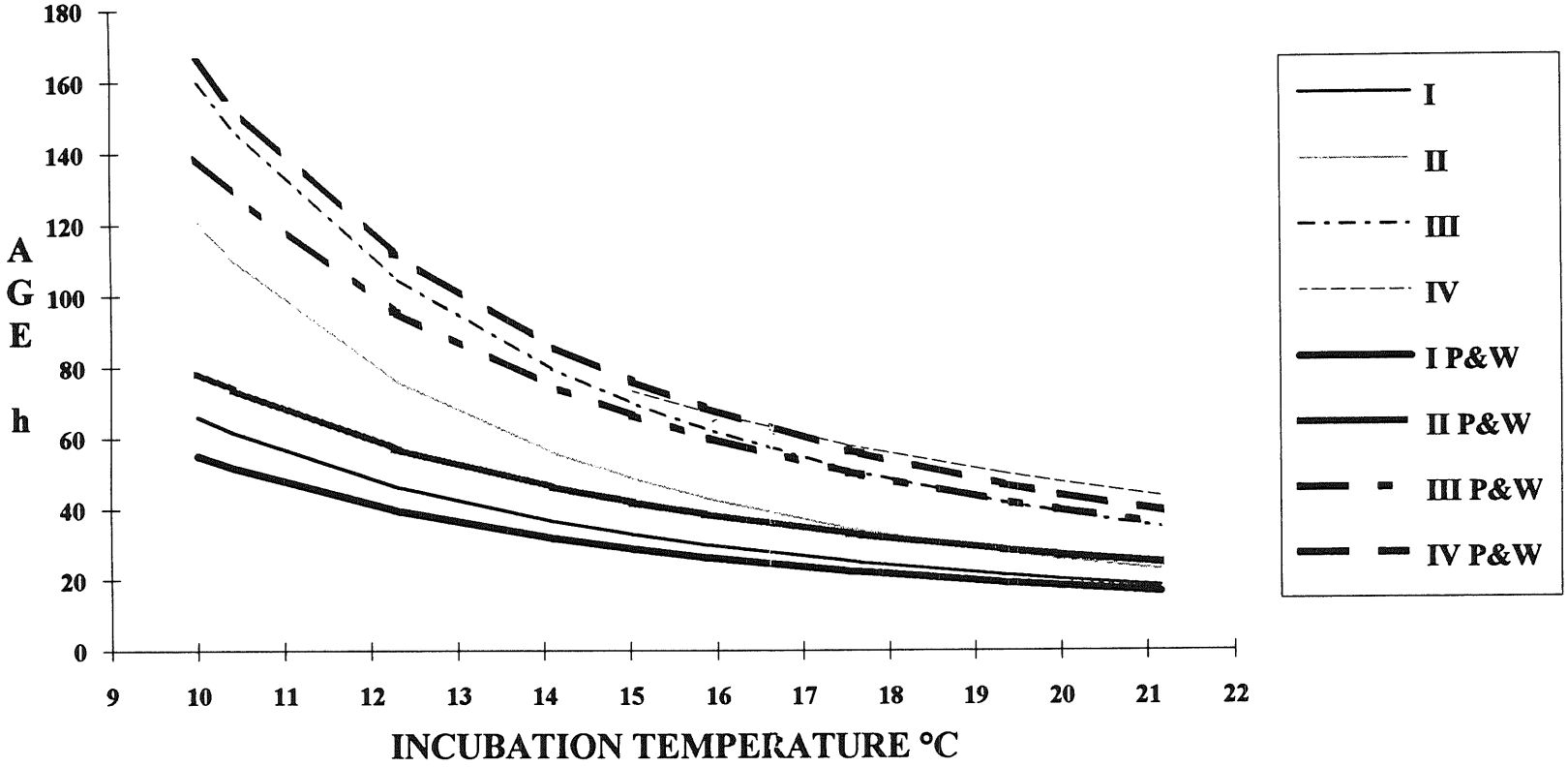


Figure 2.2 Horse mackerel egg incubation stage duration. Comparison between curves fitted by the regressions of Pipe and Walker (1987) (indicated as P&W series) and by the regressions of Motos and Muriel (WD 1994).

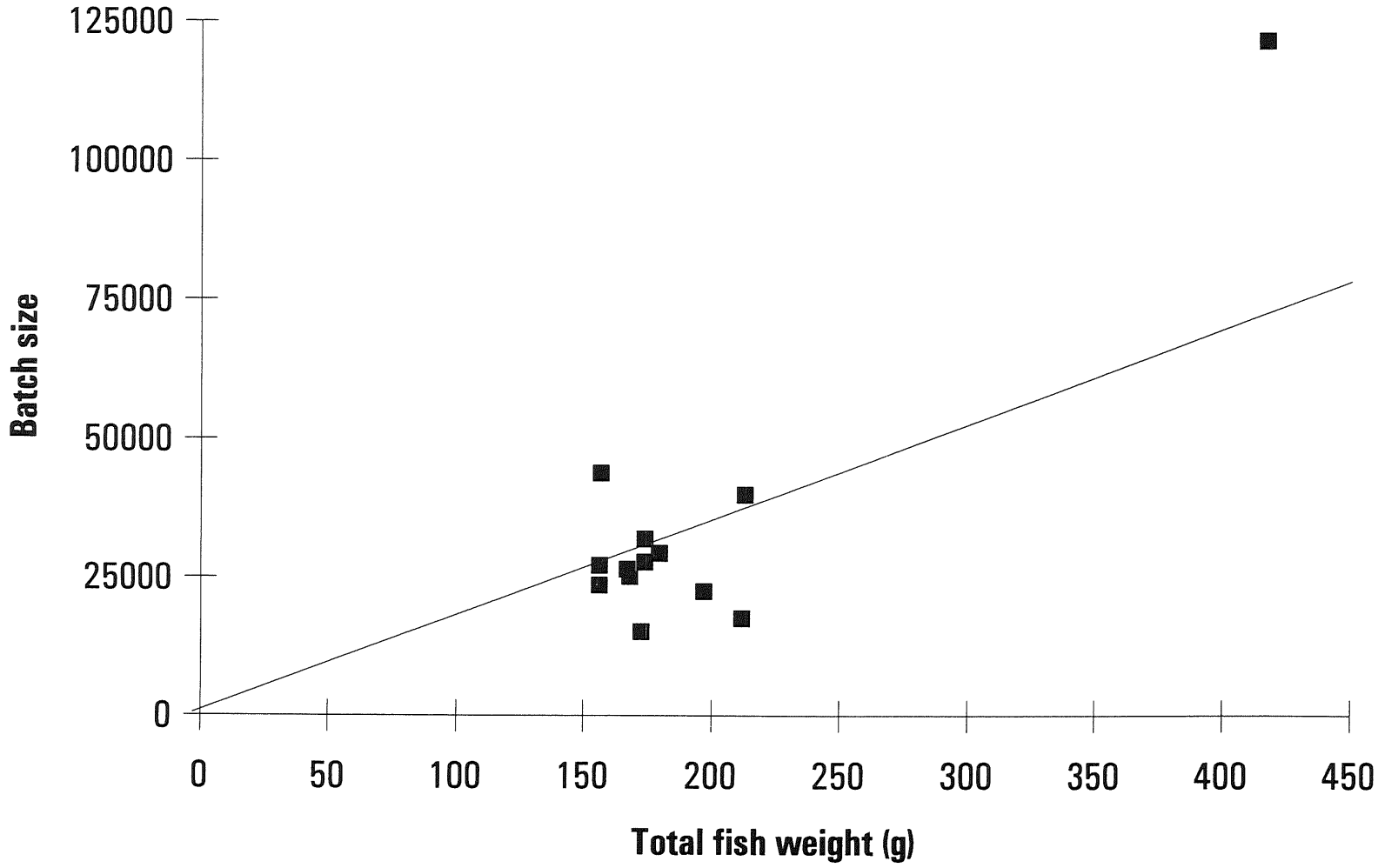


Figure 4.1 The batch fecundity-weight relationship of 196 eggs per gramme female horse mackerel in the Spanish area of Division VIIIc in 1992.

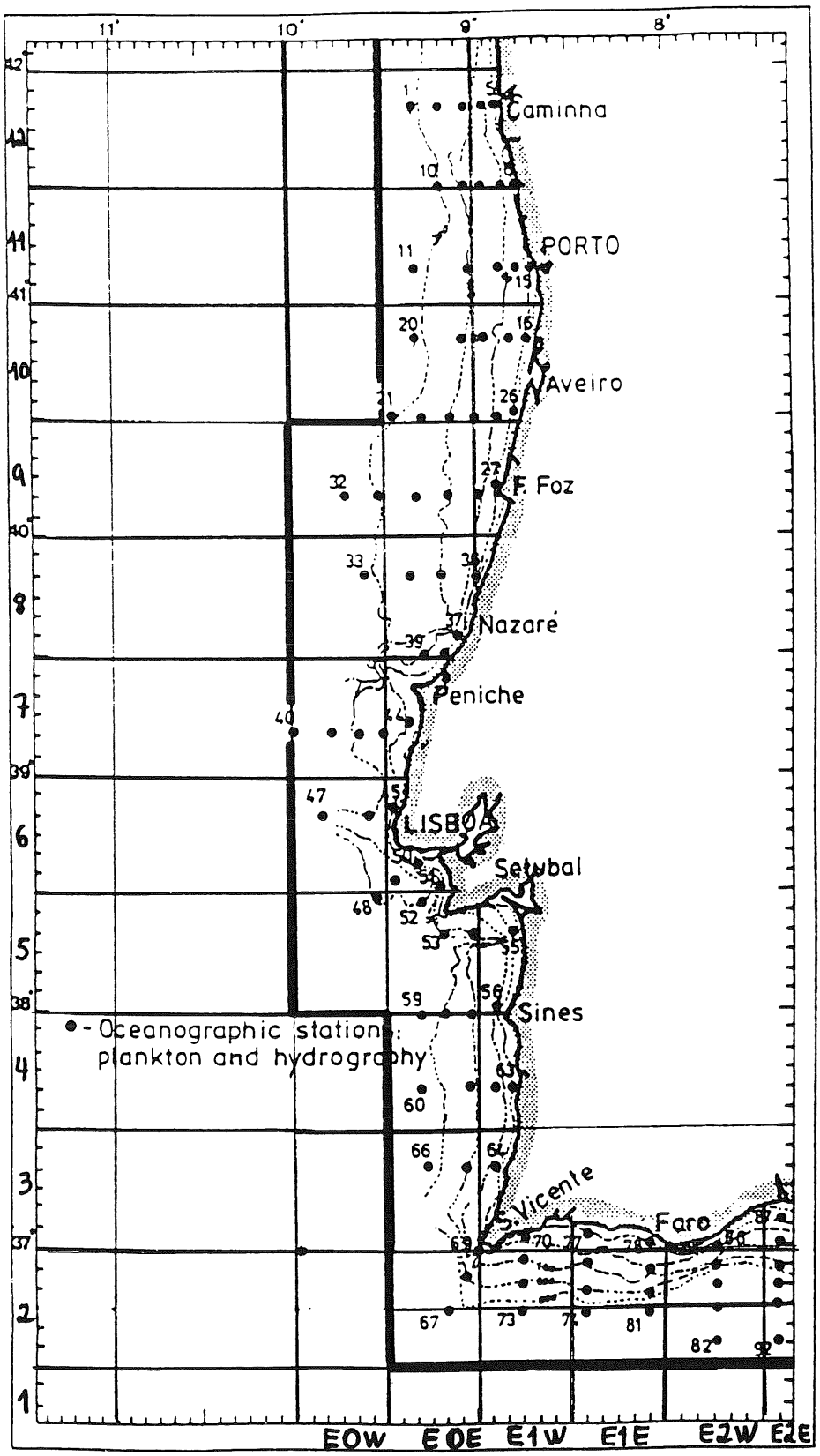


Figure 4.2 The Portuguese egg survey in February - March 1992 for estimating the daily egg production of horse mackerel.

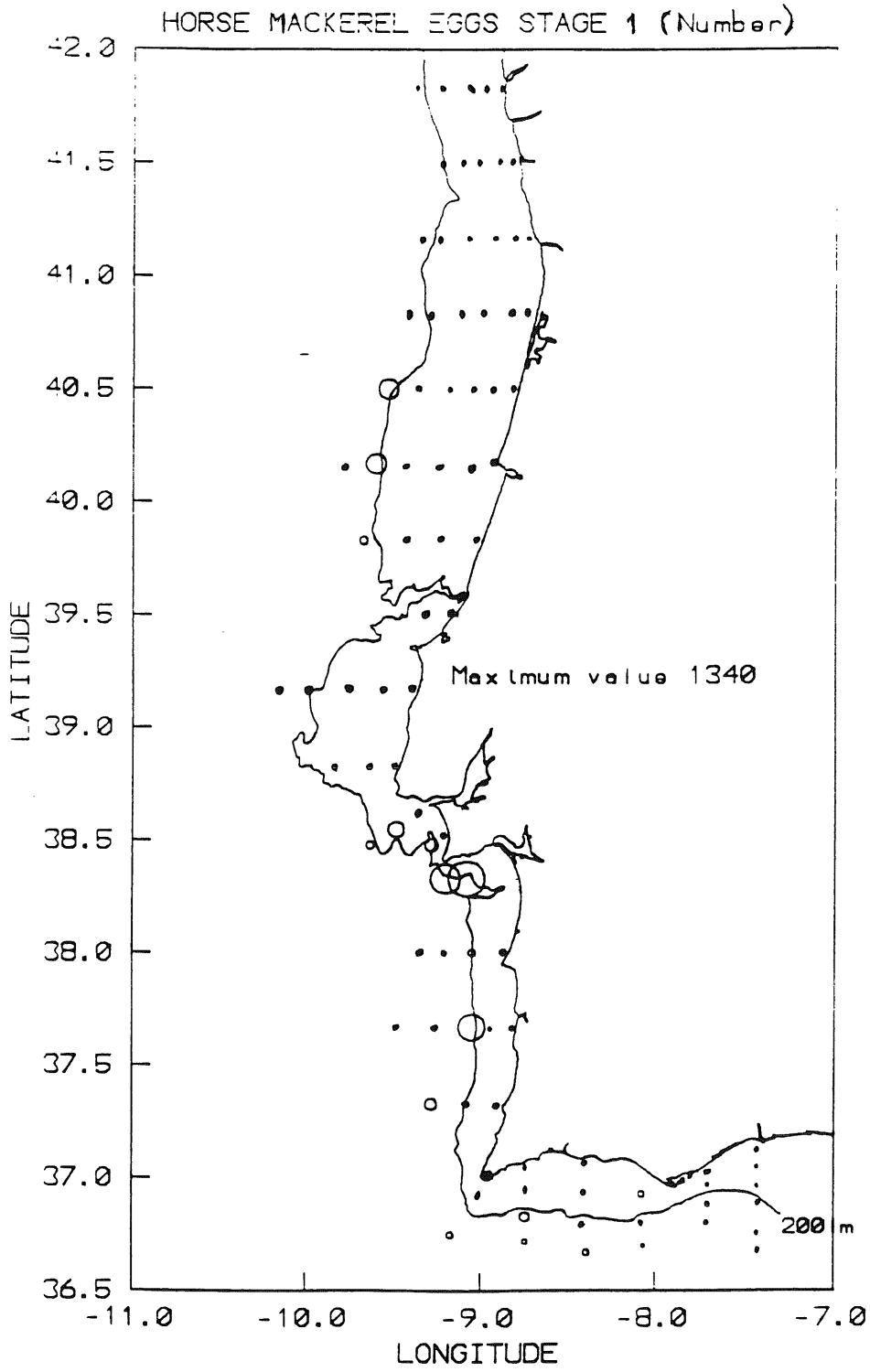


Figure 4.3 Horse mackerel stage I eggs during 14 February - 20 March 1992. Open circles are positive hauls and indicate the number of stage I eggs per m^2.

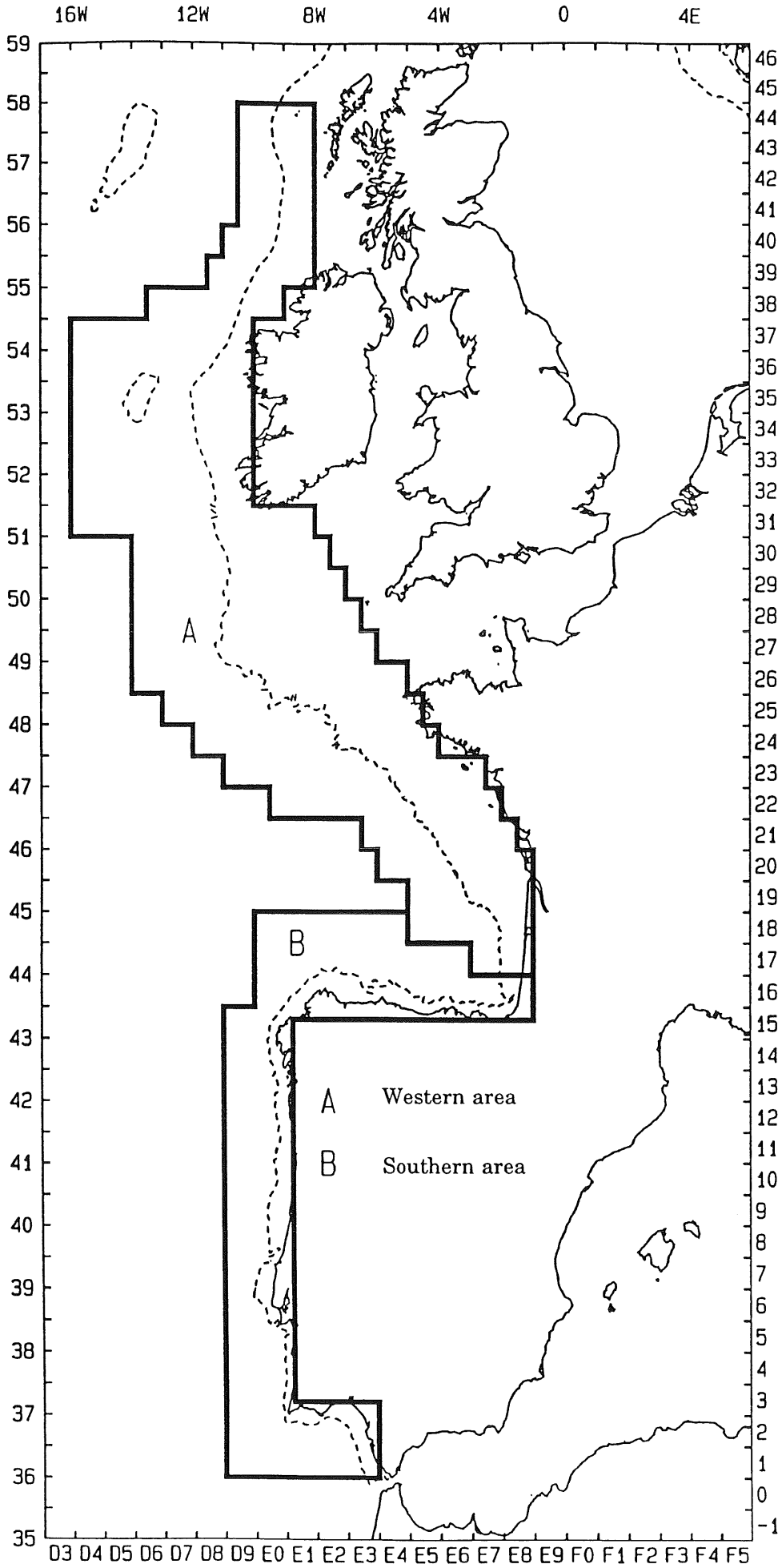


Figure 6.1 Overall sampling area for western/southern mackerel and horse mackerel spawning stocks.

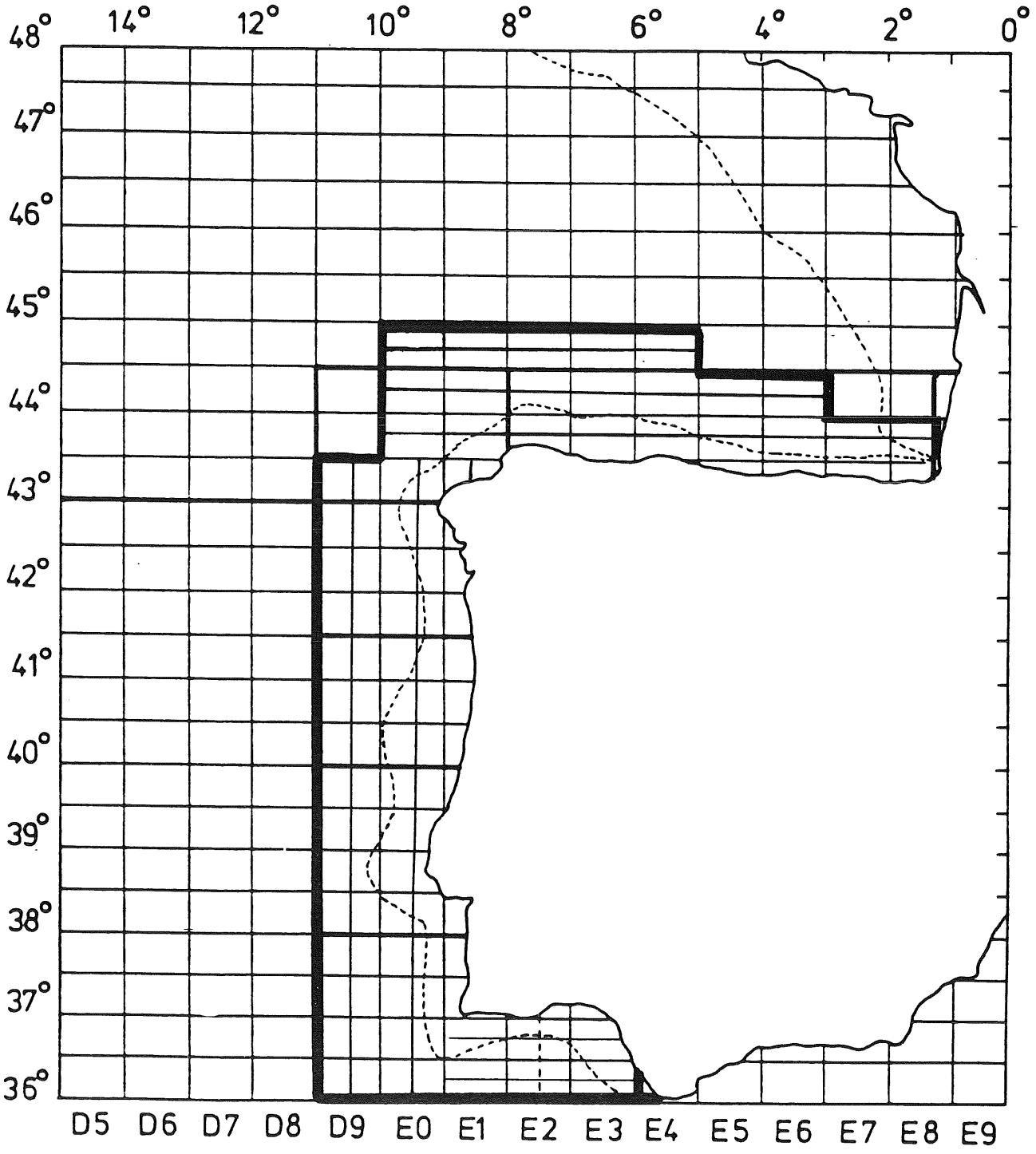


Figure 6.2 Standard sampling area of the Southern egg survey in 1995.

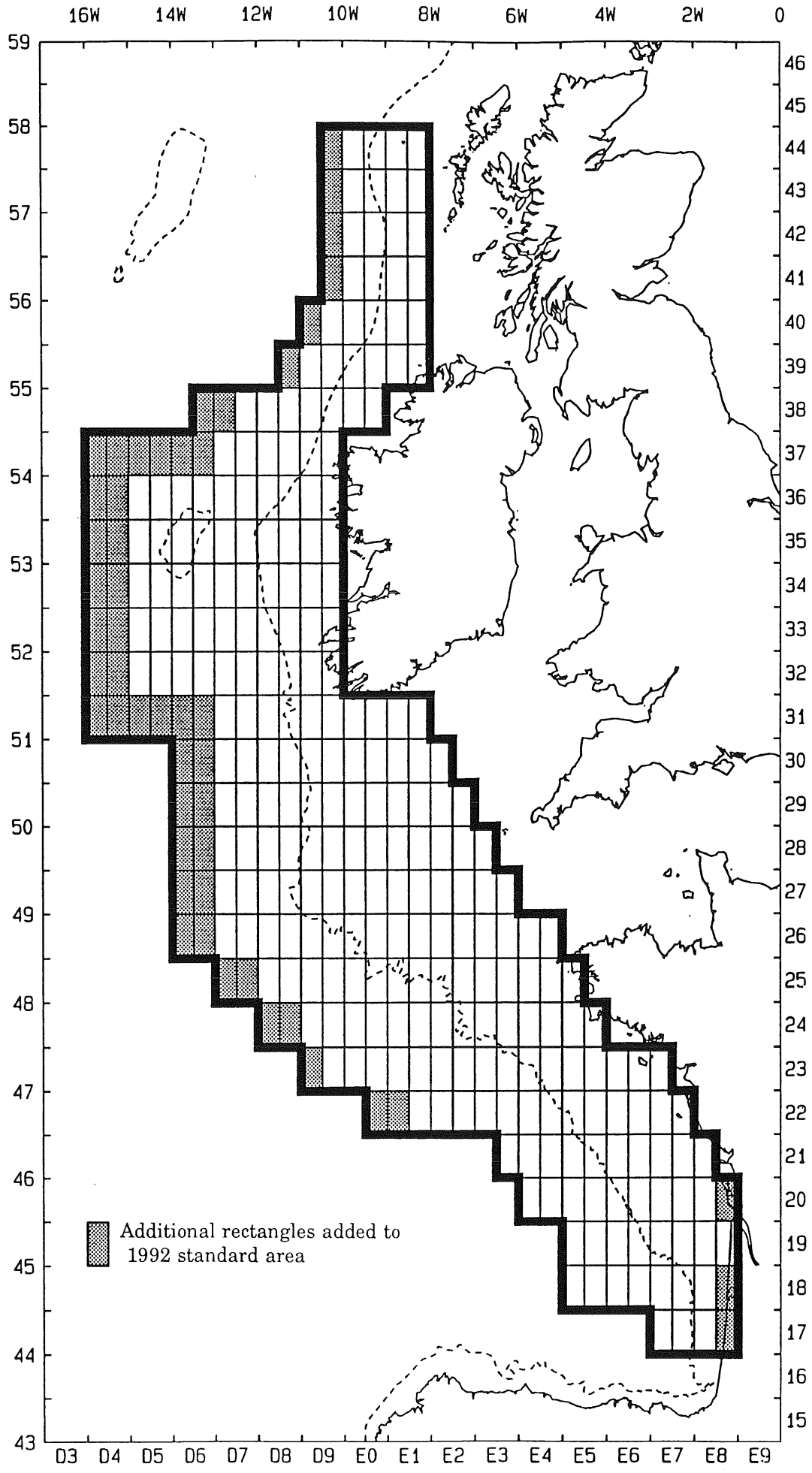


Figure 6.3 Standard sampling area for western mackerel and horsemackerel egg survey in 1995.

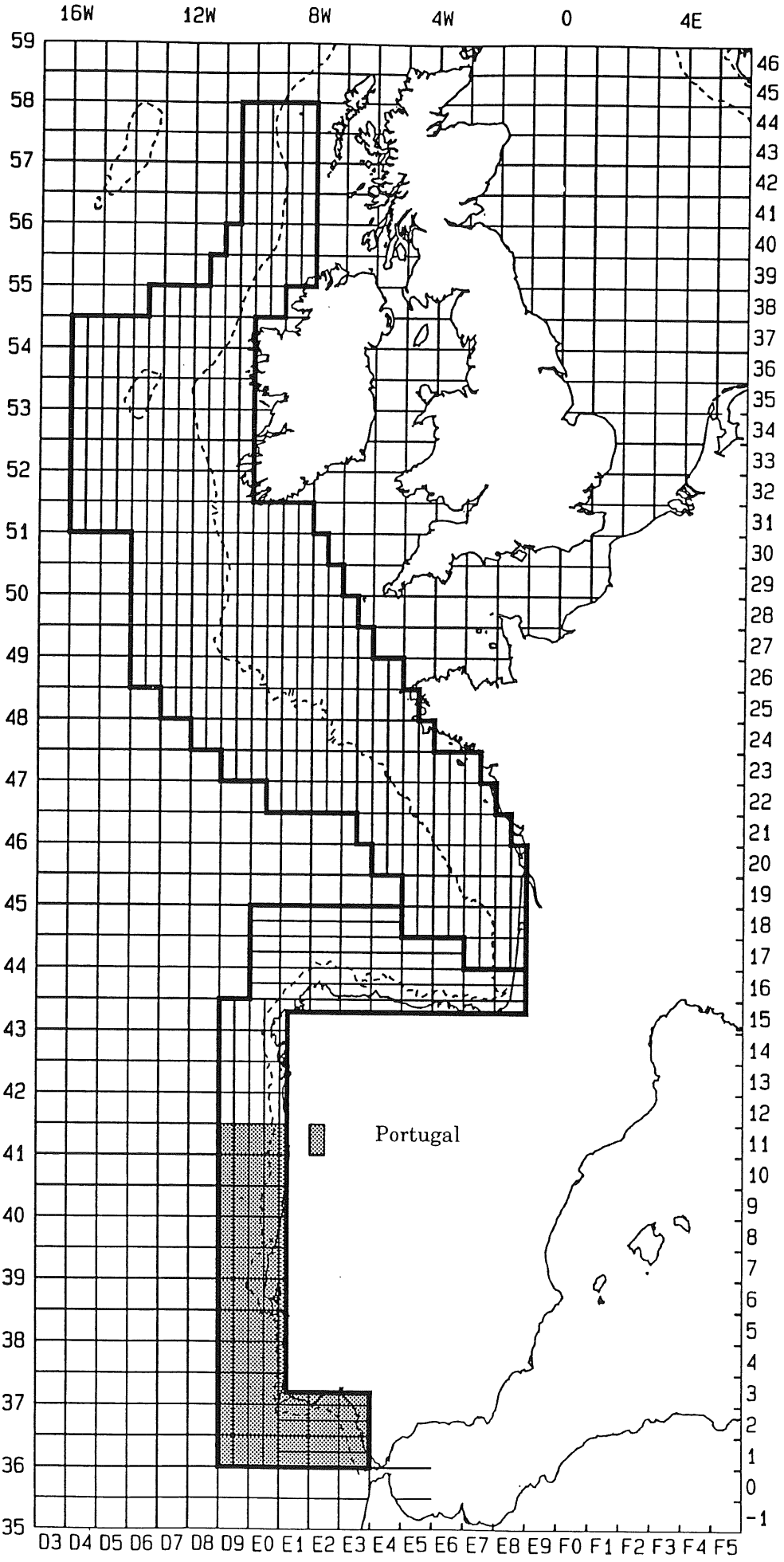


Figure 6.4 Period 1, 6-19 February 1995 proposed minimum area coverage.

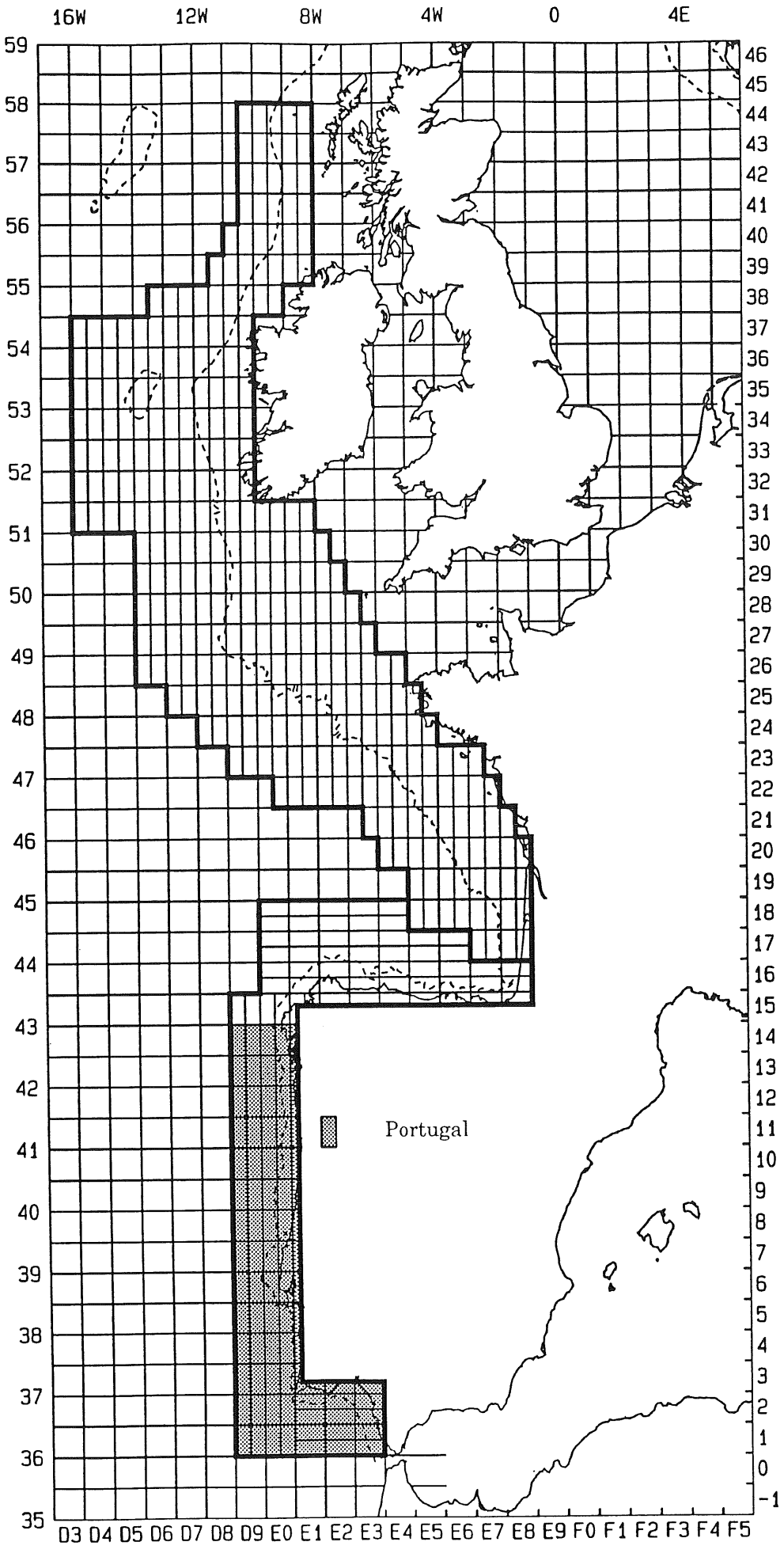


Figure 6.5 Period 2, 6-26 March 1995 proposed minimum area coverage.

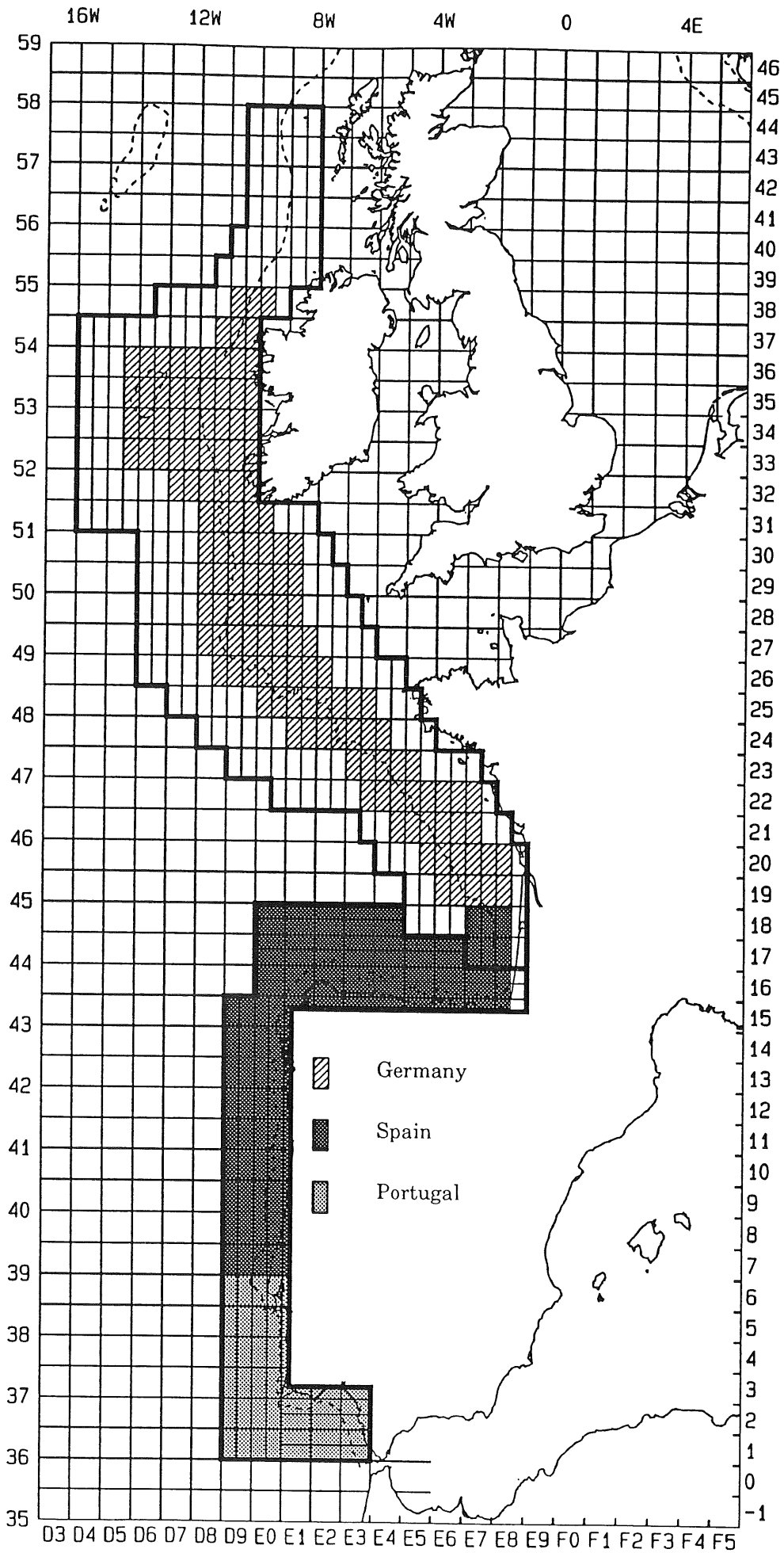


Figure 6.6 Period 3, 27 March - 16 April 1995 proposed minimum area coverage.

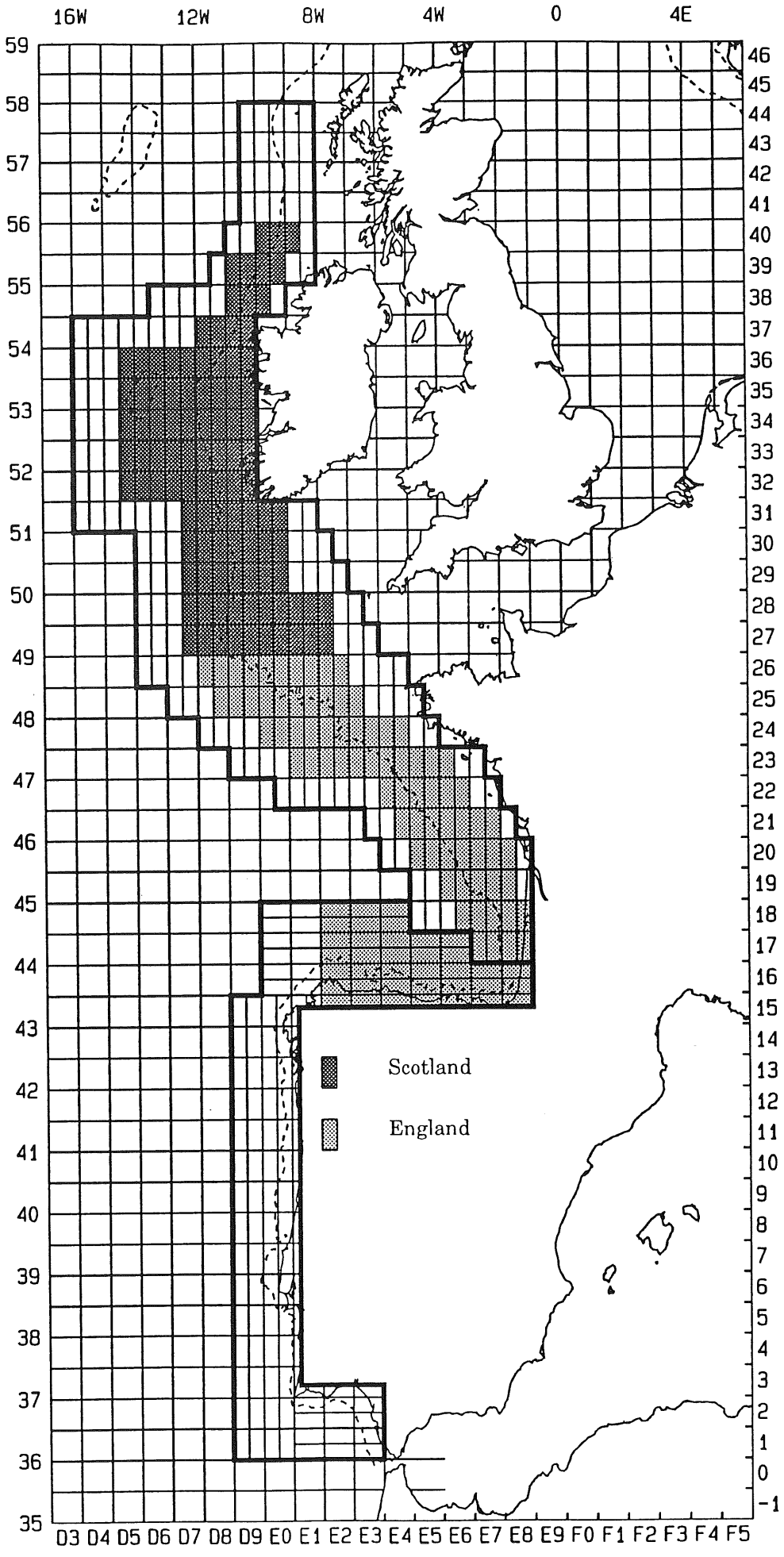


Figure 6.7 Period 4, 24 April - 14 May 1995 proposed minimum area coverage.

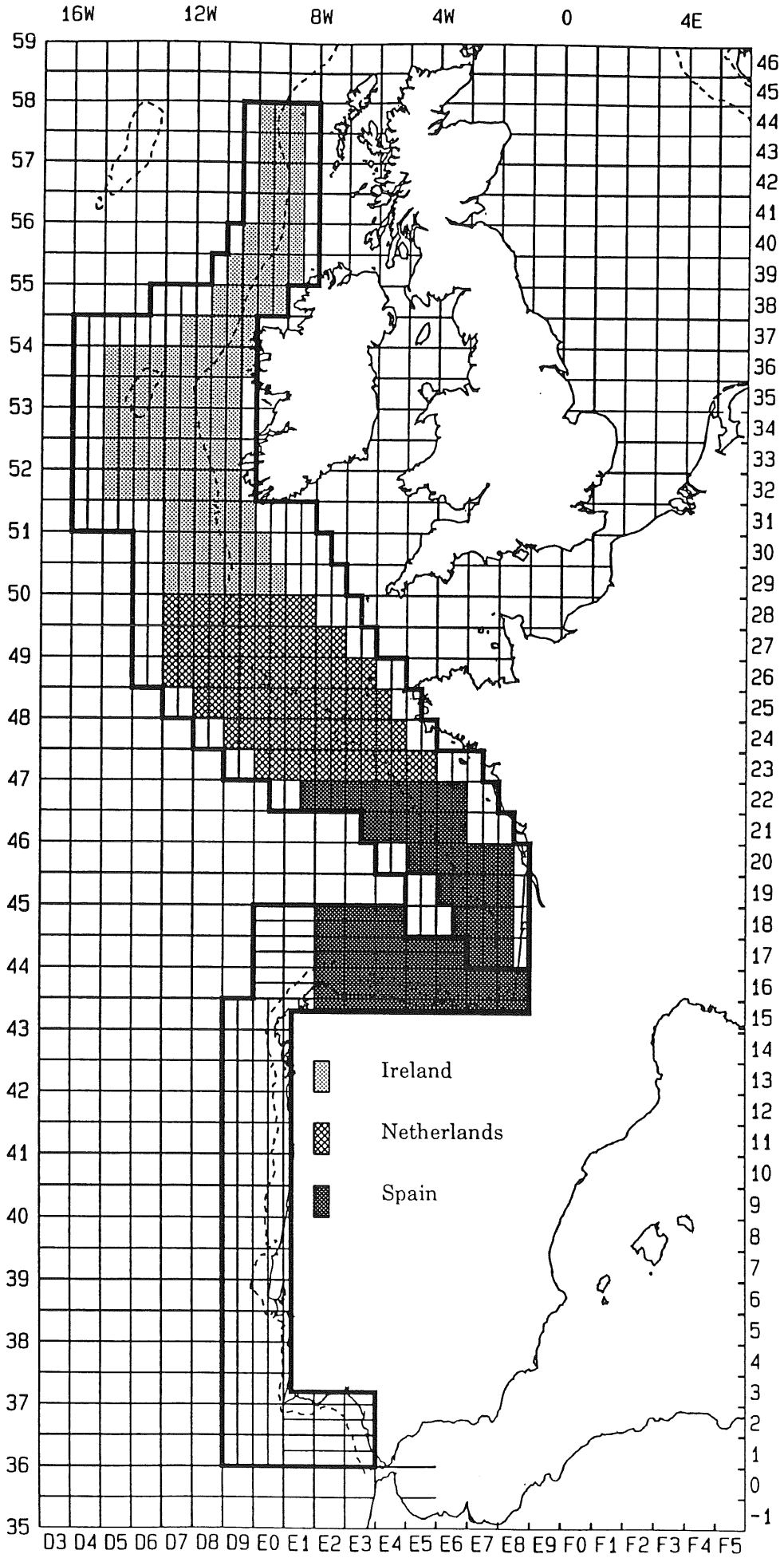


Figure 6.8 Period 5, 15 May - 11 June 1995 proposed minimum area coverage.

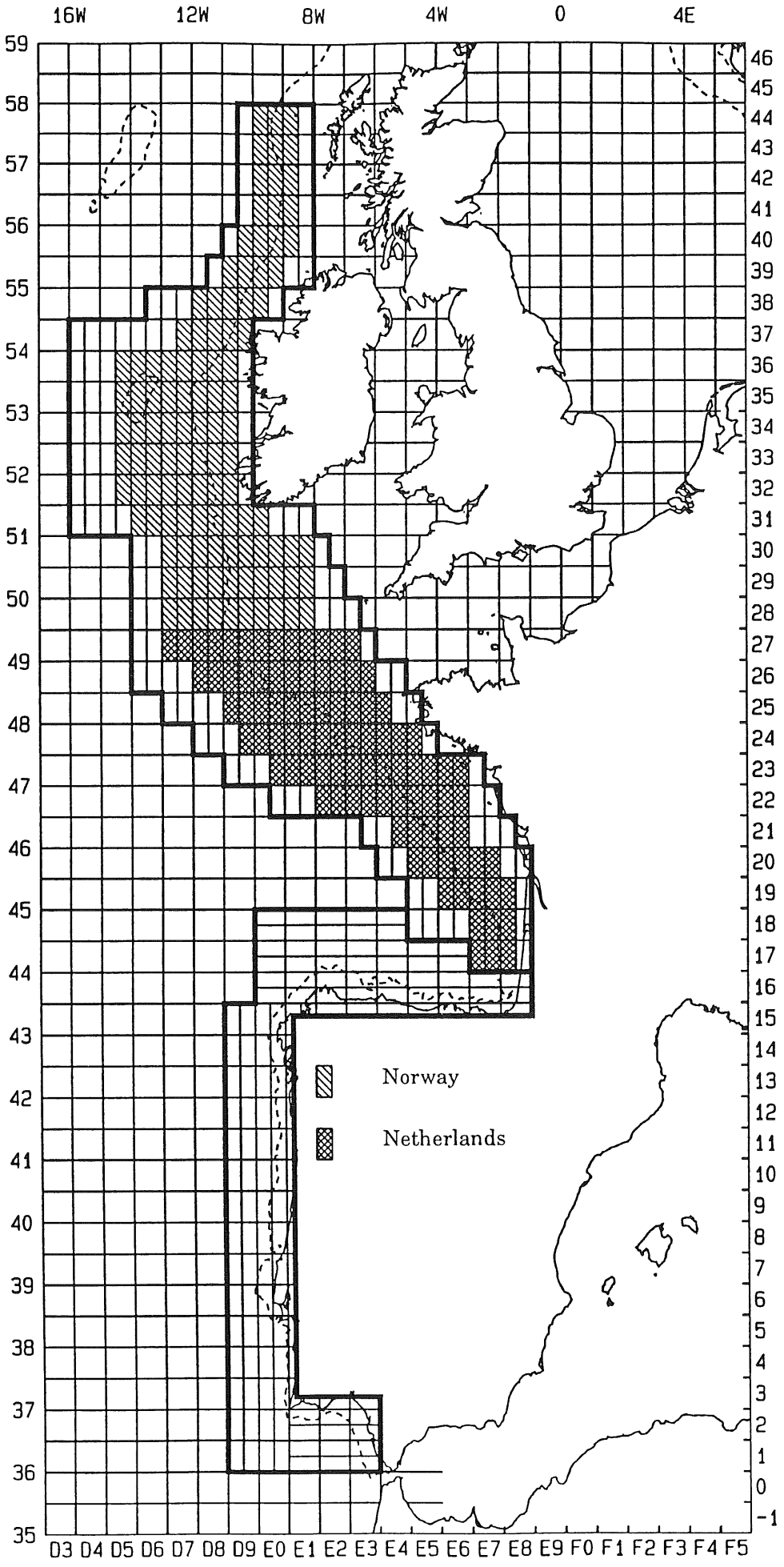


Figure 6.9 Period 6, 12 June - 2 July 1995 proposed minimum area coverage.

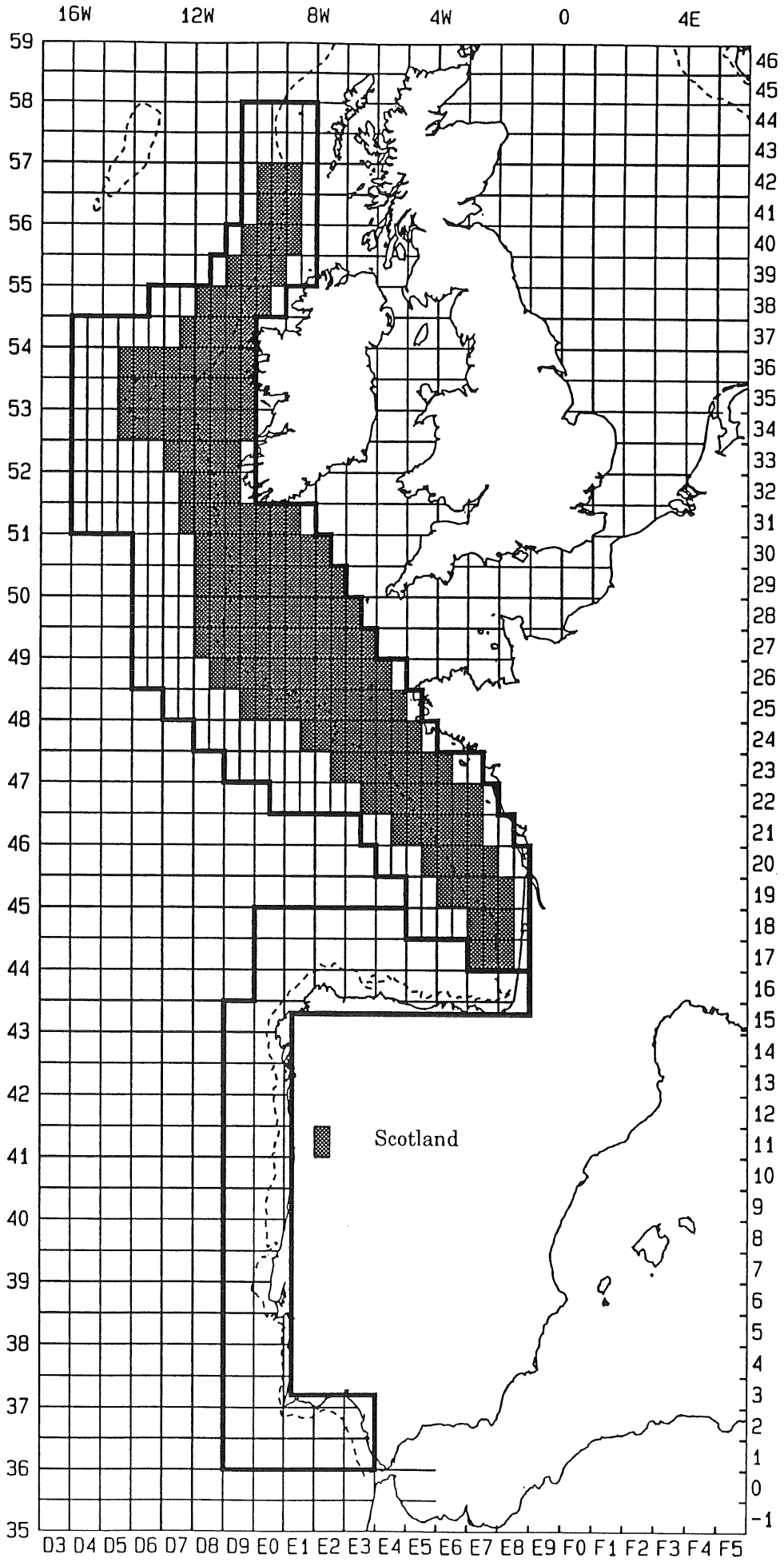


Figure 6.10 Period 7, 3-23 July 1995 proposed minimum area coverage.

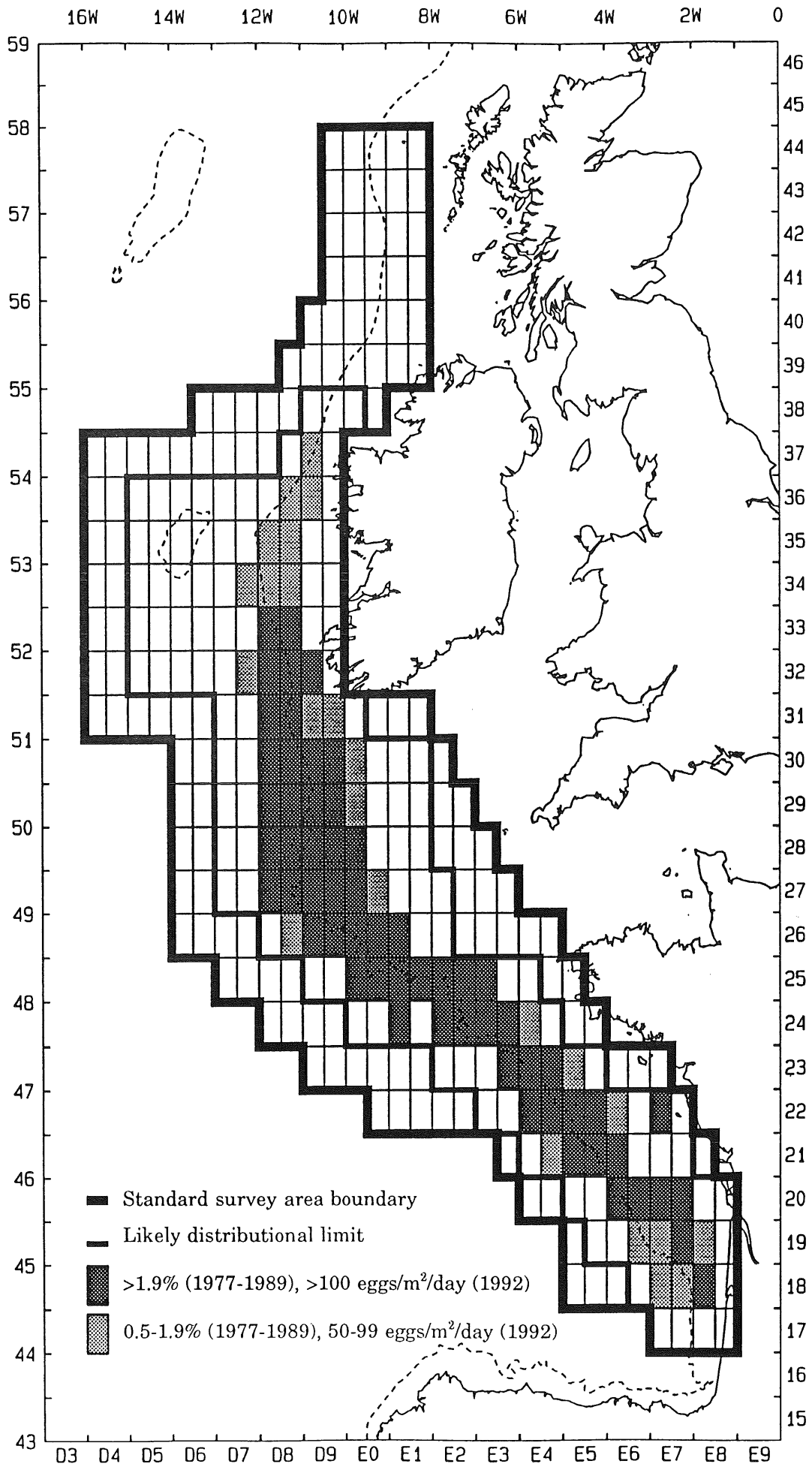


Figure 6.11 Main distributional area of stage 1 mackerel and horse mackerel eggs from previous surveys (1977-1992) and likely distributional limits for period 9 March -30 April.

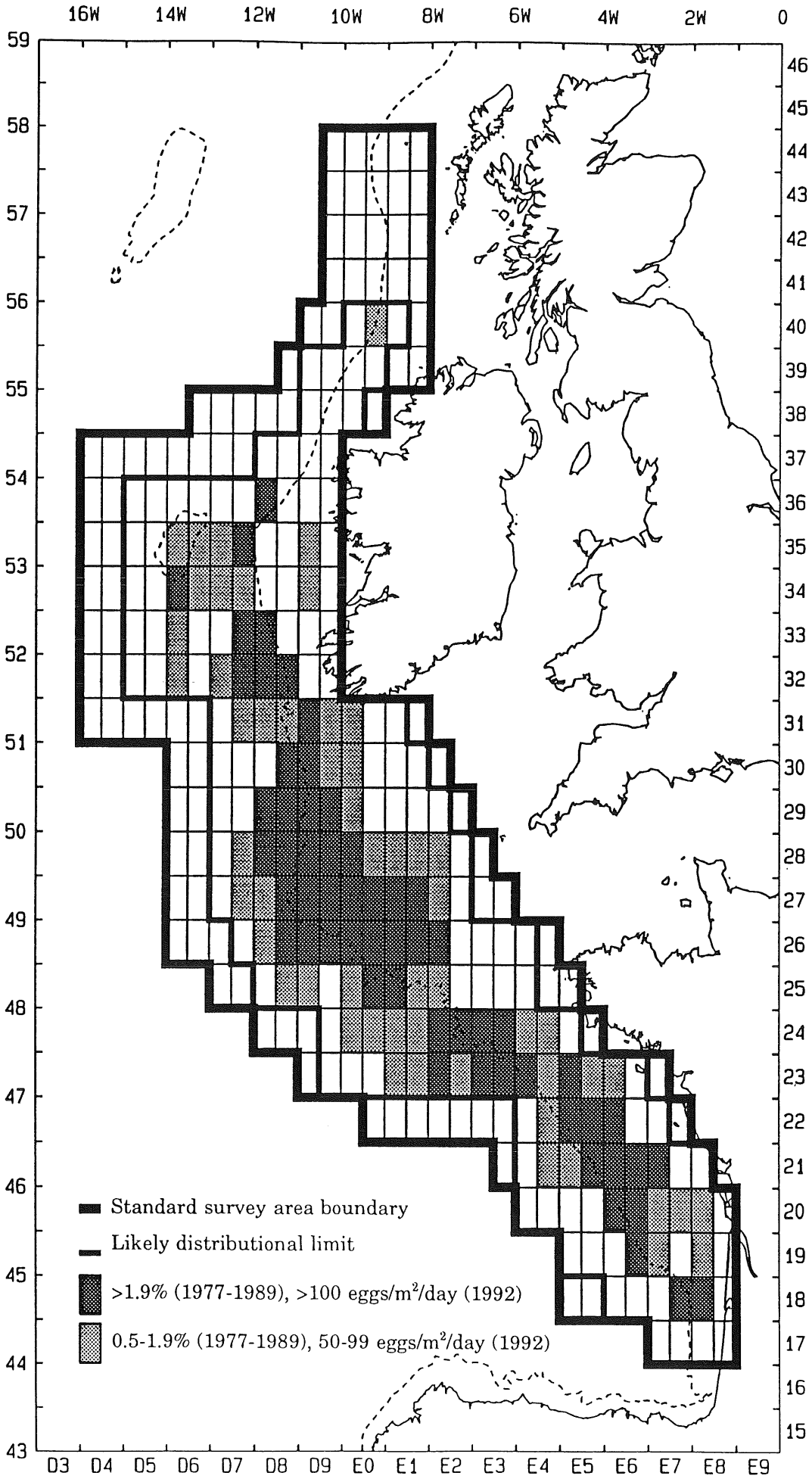


Figure 6.12 Main distributional area of stage 1 mackerel and horse mackerel eggs from previous surveys (1977-1992) and likely distributional limits for period 20 April -20 May.

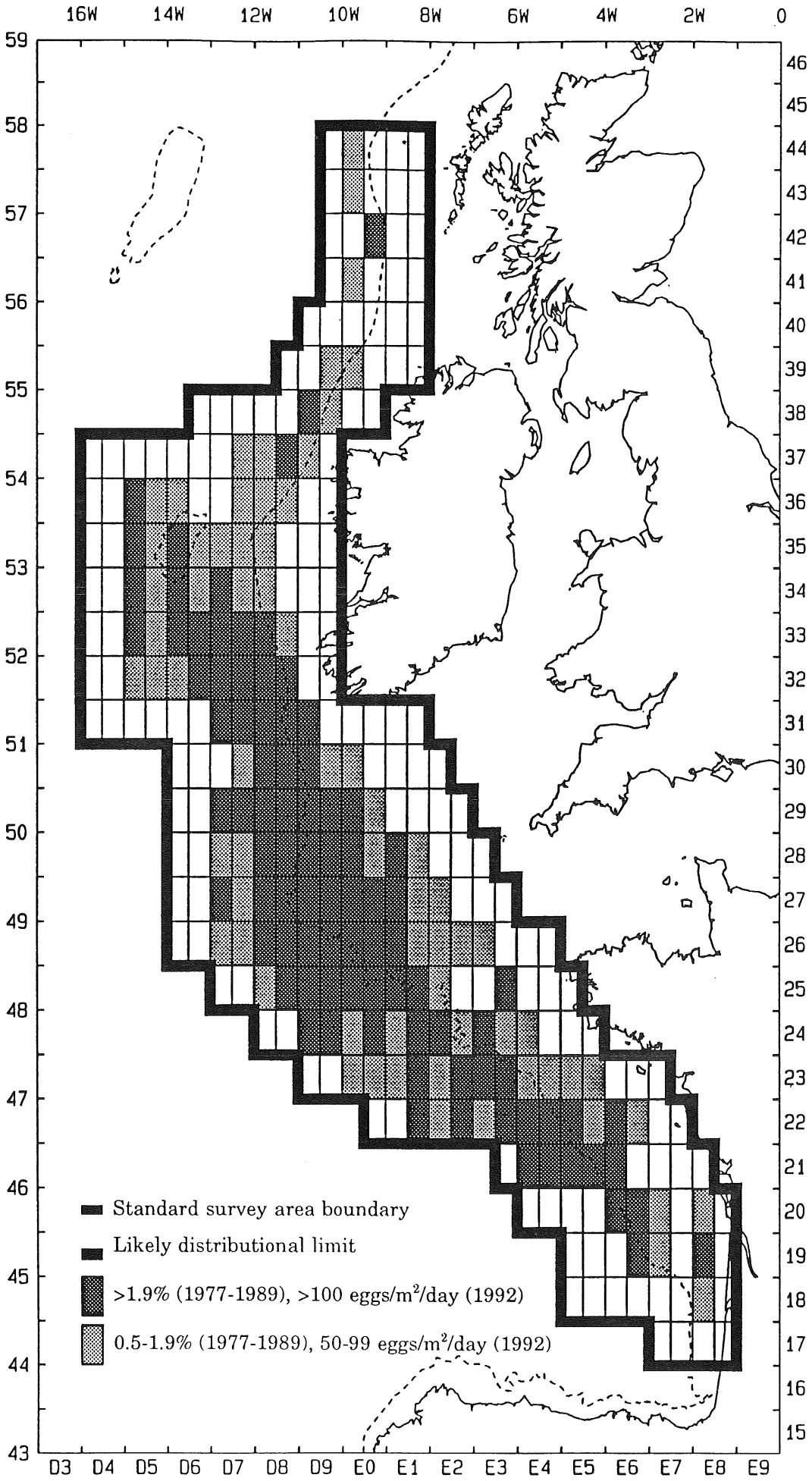


Figure 6.13 Main distributional area of stage 1 mackerel and horse mackerel eggs from previous surveys (1977-1992) and likely distributional limits for period 11 May - 17 June.

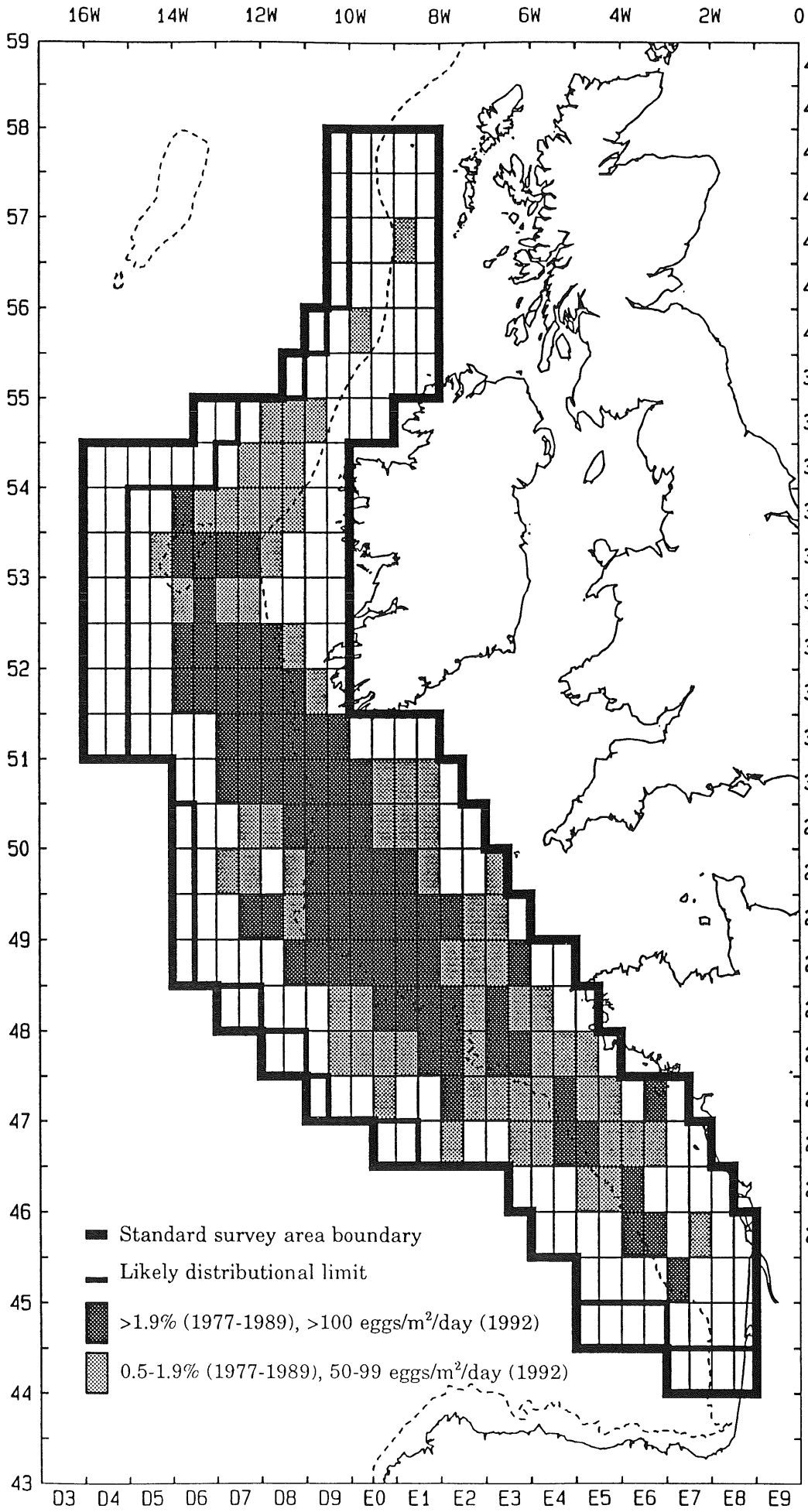


Figure 6.14 Main distributional area of stage 1 mackerel and horse mackerel eggs from previous surveys (1977-1992) and likely distributional limits for period 11 June -5 July.

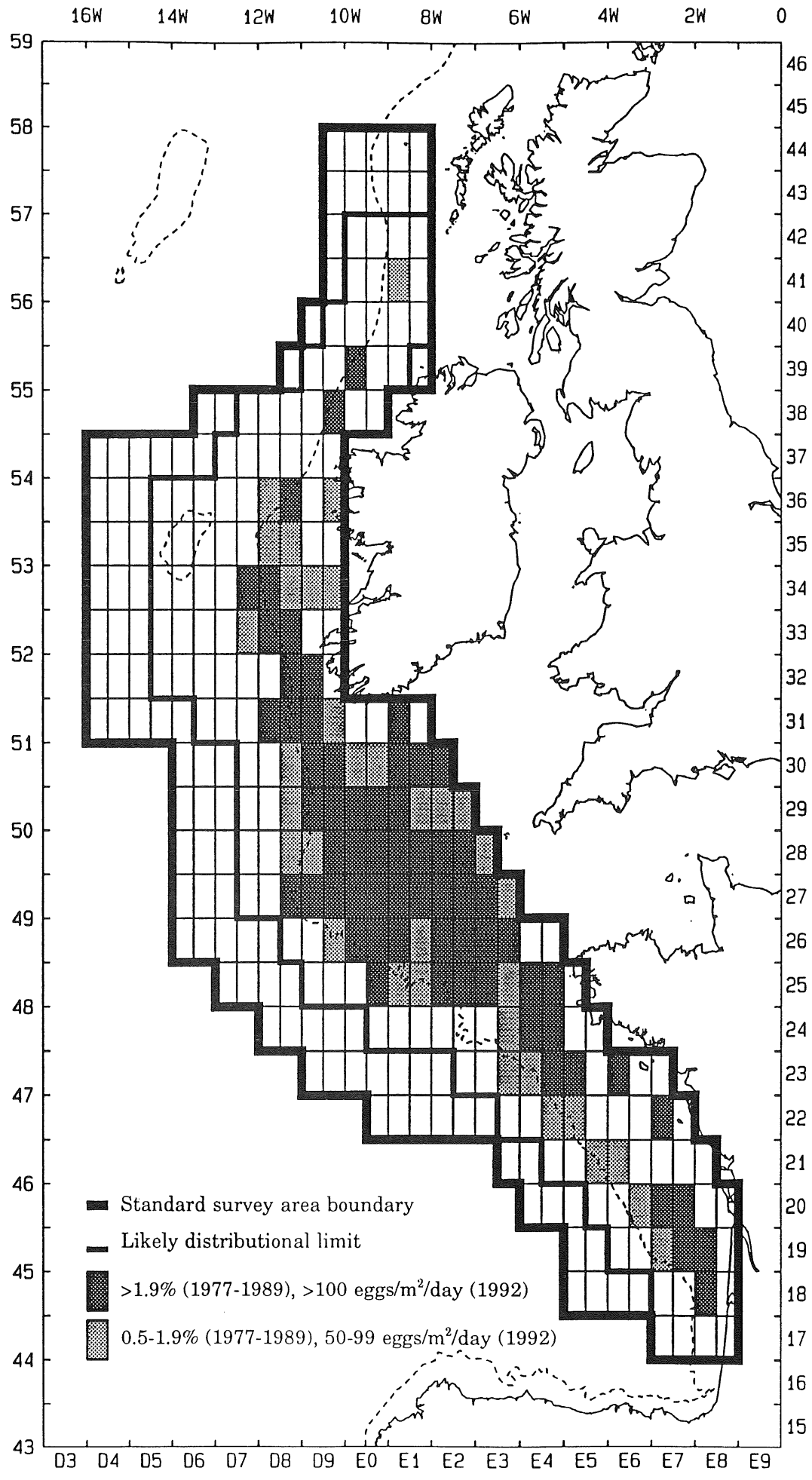
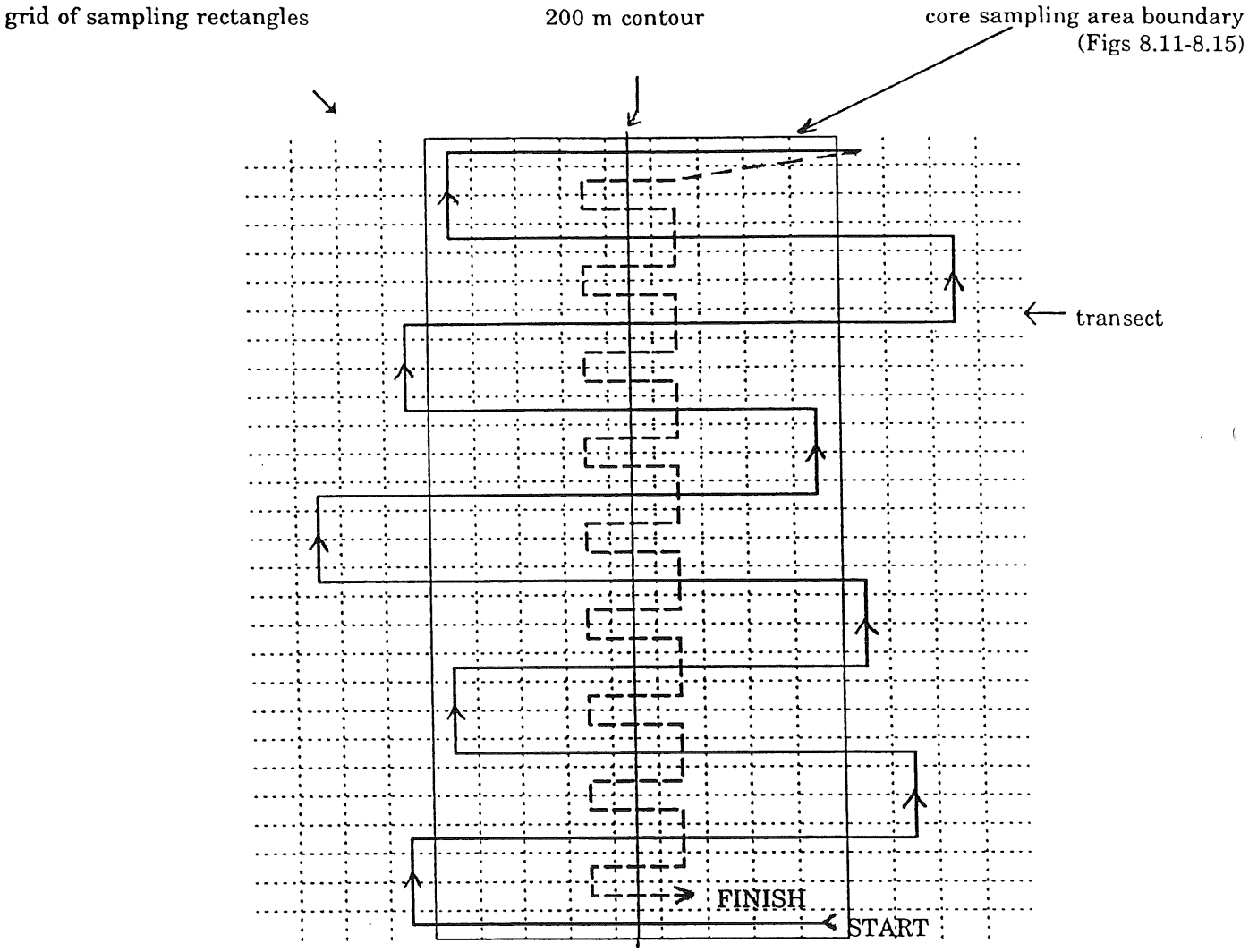


Figure 6.15 Main distributional area of stage 1 mackerel and horse mackerel eggs from previous surveys (1977-1992) and likely distributional limits for period 21 June -30 July.

Figure 6.16 Example of an adaptive sampling strategy involving sampling every third transect to limits of distribution during first part of cruise followed by sampling only central strip around 200 m contour on return



Stopping Rules for Ending Sampling in a Given Transect

1. Always sample to boundaries of core distributional area (Figs 6.11-6.15) before ending transect.
2. Stop either after one zero (or near zero) value or two consecutive low values, ie < about 20 stage I eggs of either species.

Notes: In practice eggs do not become visible until about an hour or so after fixation - roughly the steaming distance between stations - so that one extra station after a zero or two low values will always be necessary before steaming to next transect. This means that on board evaluation of egg numbers must begin three rectangles in from the outer boundary of the core area.

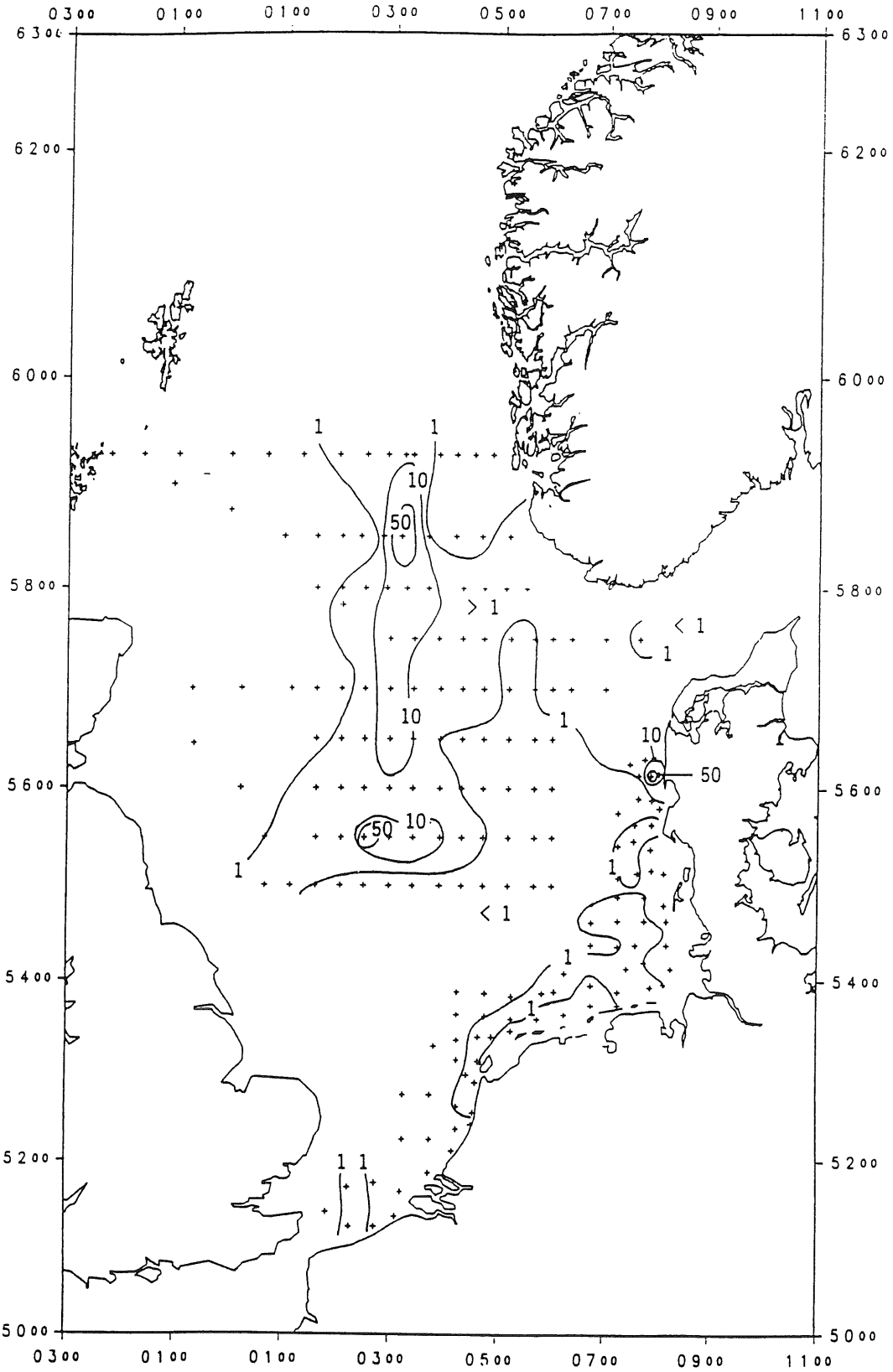


Figure 7.1 The distribution of stage I eggs/m^2/day of mackerel during the fifth coverage (17 - 30 Juny 1990) and the stations sampled (Iversen *et al.*, 1991).

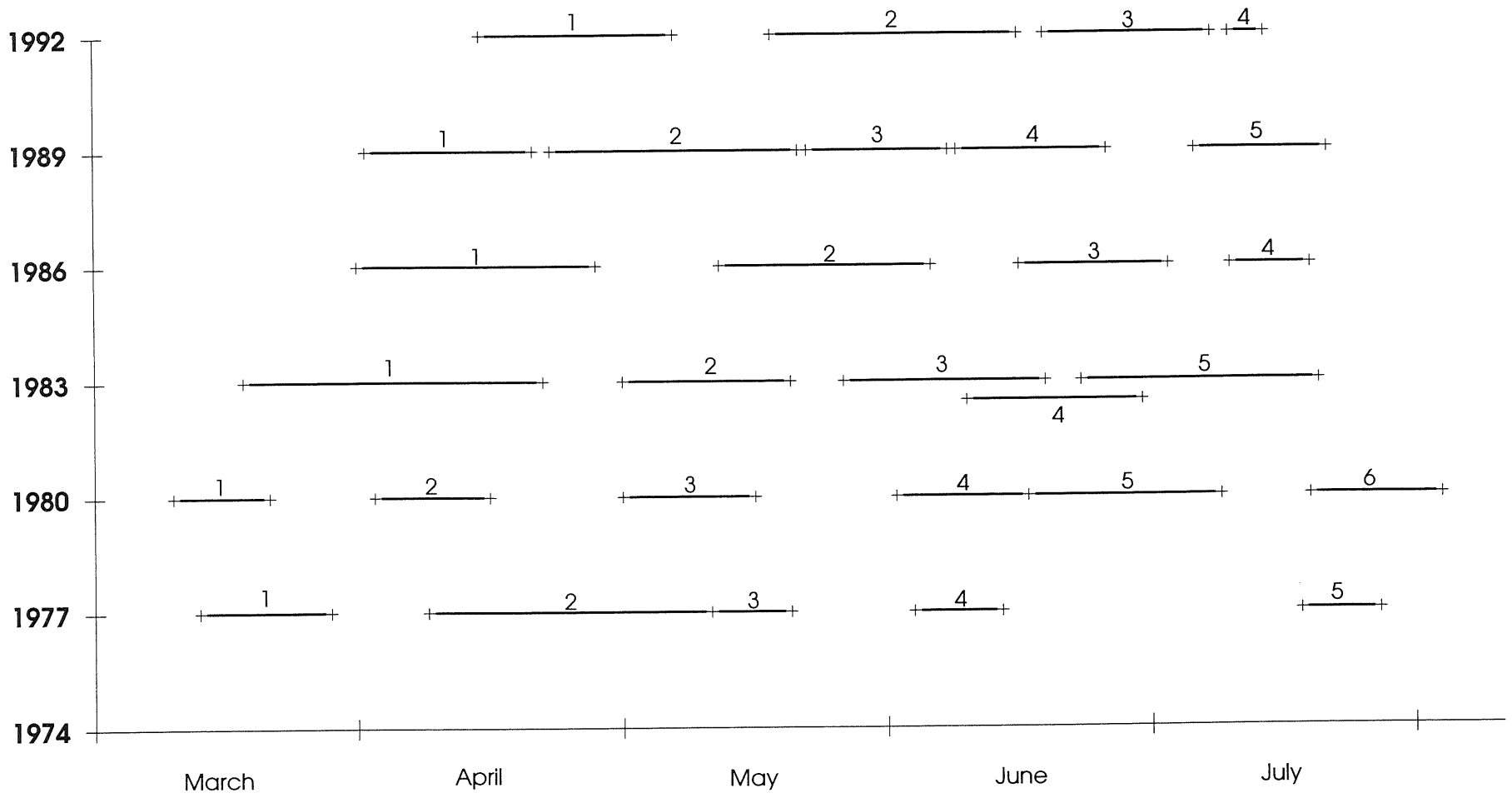


Figure 8.1 Temporal coverage of the western mackerel / horse mackerel egg surveys.