FINAL REPORT

ANALYSIS OF THE DISTRIBUTION OF PILCHARD EGGS OFF WESTERN AUSTRALIA TO DETERMINE STOCK IDENTITY AND MONITOR STOCK SIZE.

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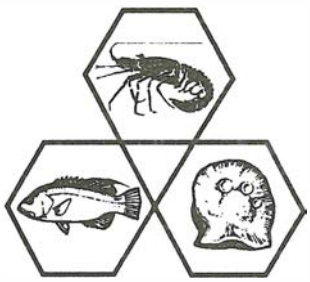

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1. Section 1 General Introduction

1.1 Background

The fishery for pilchards off the south coast of Western Australia has expanded rapidly over the past 15 years and now forms the largest volume finfish fishery of this state with a catch value in the vicinity of 6 million dollars. Information on both the number and size of pilchard stocks along this coast was required to determine if the levels of exploitation in the established fisheries were appropriate and whether the expansion into the Bremer Bay and Esperance regions was justified.

Obtaining estimates of stock size and the pattern of distribution of schooling pelagic fish is difficult because their behaviour makes fishery dependent methods (eg. CPUE) unreliable estimators. Catchability is not constant with stock size and alterations to migration patterns will influence catch rates in an area despite no changes in the actual abundance having occurred. Consequently, many stocks of pelagic fish are now monitored using the fishery independent techniques of egg/larval abundance and distribution indices. These can either be a relative index, such as the area of spawning, or alternatively, an absolute measure of spawning biomass can be calculated using the daily egg production method. The latter, however, also requires sampling of the adult fish to determine batch fecundity and spawning frequency.

1.2 Need

The application of planktonic based techniques to the pilchard stock of Western Australia commenced in July 1991 when an initial plankton survey (FRDC Study 91/24) of the pilchard egg distribution was made covering the region between Bremer Bay and Pt D'Entrecasteaux. The results of this survey clearly showed that the distribution of pilchard eggs was not uniform along the south coast. There were concentrations of Day-one eggs in the region off both Albany and Bremer Bay, the areas where the majority of fishing occurs. Utilising the pattern of egg abundances, along with the concurrent samples of adult pilchards, allowed a preliminary estimate of the spawning biomass off the Albany region to be calculated. It also enabled the pattern of adult abundance to be compared with predictions of stock distribution generated by a computer model (see Fletcher, 1992; Fletcher *et al.,* 1994).

The distribution of eggs can be used to help delineate pilchard stocks. Thus, to the west of Albany, virtually no eggs or traces of fish were found suggesting that little spawning and indeed few pilchards were located in this area. This suggests that the pilchards on the west coast (ie. those caught at Fremantle) may be a separate stock from the south coast fish. However, this pattern of few eggs west of Albany coincided with the main influence of the Leeuwin current which was flowing close to this area of the coast. It was, therefore, necessary to repeat these tows, and extend the sampling around to the west coast to determine if this pattern is a consistent feature or merely a transient effect of local oceanographic conditions.

To the east of Albany through to where sampling ended at Bremer Bay, increasing numbers of larvae were found in the samples, probably a result of larval drift. This suggests that the stocks of pilchards may be mixed quite widely during their planktonic phases. The relationship between the fish in the Albany/Bremer Bay areas with those at Esperance was unknown and hence required the sampling regime to be extended. Different management strategies would result depending upon whether the areas contain the same or separate adult stocks. It would also be valuable to determine the relationship of the Albany, Bremer Bay and Esperance stocks with stocks further to the east in the Great Australian Bight (GAB) and South Australia (SA).

1.3 Objectives

Initial

(i) Document the distribution of eggs and larvae of pilchards along the south and south west coasts of Western Australia during the critical winter season to determine the relationship of spawning at Esperance with the more western sites of Albany and Bremer Bay.

(ii) Determine the relationship of south and west coast pilchard stocks.

(iii) Examine inter-annual variation in the distribution and abundance of eggs in relation to changes in catch rates by the fishery and variations in hydrological conditions, particularly water temperature.

(iv) Provide data inputs on the distribution and abundance patterns of pilchards for the computer simulation model built to assess the state of the fishery.

Additional since project began

(v) Calculate biomass estimates for some of the regions using the daily egg production method.

(vi) Determine the relationship between stocks of pilchards in WA with those in the GAB and SA

2. Section 2: General Methods

2.1 Sampling

Samples were collected over a three year period, with 8 separate surveys completed. The area covered in total by all 8 surveys extends from Fremantle in Western Australia to Adelaide in South Australia (Fig. 2.1). The dates, areas covered, the type of nets, the profile of the tow, number of stations sampled and vessels used varied for each trip and are detailed in Table 2.1.

For ease in the recording of samples on board the boat and for their later analysis, the coast line was divided into a number of blocks based on their geographical location. The boundaries of each block are shown in Fig. 2.2. Sampling stations were also coded depending on whether they were inshore, mid-shore or offshore.

2.2 Nets Used

Six types of nets were used during the study. All vertical and oblique tows were conducted with a bongo-net arrangement. Two types of bongo nets were used (Fig. 2.3) - either the standard type or a modification of the CalVET net type (Smith *et al.*, 1985). The standard bongo arrangement was fitted with either 500 μm mesh nets on both rings, or a 500 μm on one ring and a 300 μm mesh net on the other. Both nets used on the CalVET arrangement were 300 μm mesh.

The nets used surface tows were essentially conical but with 1 x 1 m and 0.5 x 0.5 m square frame openings. The nets fitted to both frames had 1000 μm mesh.

The last type of net is the "EZ net". This was used to conduct tows at specified levels in the water column aboard the *R.V. Franklin.* The EZ net has a multiple opening and closing arrangement whereby up to 10 nets of 1.0 m^2 mouth area can be deployed in a single tow. An on board computer relays signals to open and close the nets, allowing the duration and depth of each sample to be individually controlled. The depth, temperature, salinity and tow angle were displayed and digitally recorded throughout the tow.

The EZ unit was deployed astern with all nets in the 'closed' position and allowed to sink up to 100 m depth, or 10 m from the bottom. It was then raised to the desired depth and the first net mouth opened. The net would sample at the specified depth for 2 minutes and was then closed. The unit was then raised to the next depth where the next net would be opened. Up to a total of 5 depths were sampled at any one station.

All bongo and surface nets had cod-ends made from PVC plastic with two round, meshed windows cut into the side. All nets were fitted with a mechanical flowmeter (*General Oceanics* or *Rigosha*) to measure the amount of water filtered.

The tow ropes or wire were attached to the nets with a swivel and shackle. A 6-9 kg weight was attached to bongo nets during vertical and oblique tows with heavier

weights being used in rough weather. On the standard Bongo nets, the weights were attached to the net frame near the mouth of the net while on the CalVET nets the weights were attached to a cross bar which extended between the two cod-ends (see Fig. 2.2). The latter arrangement ensured that the CalVET nets only sampled when being hauled to the surface, whereas standard Bongo nets sampled a small proportion while sinking with the majority of the water filtered whilst being hauled. During surface tows, either a weight or standard depressor were used to keep the net submerged.

2.3 Towing Methods

All tows conducted on vessels other than on the *R.V. Franklin* used nylon rope marked at 5 metre intervals. The nets were deployed over the side of the vessel to a depth not greater than 70 metres or to within 3 metres of the bottom and then winched to the surface at a speed of approximately 1 metre per second, thereby obtaining a vertical profile of the water column.

All stations conducted aboard the *R.V. Franklin* used a wire cable attached to a winch. For the vertical tows, the nets were lowered to a depth of not more than 100m or to within 3 metres of the bottom and winched to the surface at 1 metre per second. For oblique tows, the net was lowered to 150 m or 5m from the bottom whilst the boat moved ahead at 3 knots. The net was winched back to the surface at 1 metre per second. Surface tows were conducted by lowering the net such that the mouth

sampled within the top 1 metre of the water column, occasionally breaking the surface. It was towed in this position for ten minutes.

All nets were hosed down and the contents of the cod-ends placed in a buffered solution of 5% formalin and seawater. The contents of 1000 μm surface nets were partly sorted prior to fixation to remove larger pilchard larvae before being placed in the formalin solution. These larger larvae were placed in 70% denatured ethanol.

2.4 Temperature and Salinity Measurements

Temperature measurements were by one of three methods depending on available resources. In the majority of surveys the sea surface temperature was obtained by scooping a bucket of water from the sea surface and within it placing a mercury thermometer. The January 1994 and July 1994 (west of Albany) surveys utilised a TPS *LC87* electronic thermometer deployed over the side of the vessel. All temperatures obtained aboard the *R.V. Franklin* were taken from measurements of the onboard CTD.

Salinity measures obtained aboard the *R.V. Franklin* were obtained from the onboard CTD. All other salinity measurements were obtained from water samples taken from the sea surface with a plastic bucket. The samples were taken back to the laboratory and salinity readings were obtained using a *Yeokal 620* (July 1993 and January 1994 surveys) or a *WTW LF320*.

2.5 Sorting

All samples were transferred to a solution of 3% buffered formalin in seawater within a few weeks of capture. Any large volume of gelatinous material was removed prior to the measurement of displacement volume. Samples were sorted under a dissecting microscope. All pilchard eggs and pilchard larvae were enumerated, staged and placed in 3% buffered formalin or 70% ethanol respectively. All other fish larvae were enumerated and placed in 70% ethanol.

2.5.1 **Egg Staging**

The ability to accurately stage pilchard eggs is affected by a number of factors. These include the initial state of the embryo (whether it was dead or not prior to capture), damage during capture and the effect of preservation and storage (formalin tends to shrink the embryo with time and the tissue also becomes more opaque). These all impact upon the ease with which pilchard eggs can be staged. Thus, the condition of the eggs are often less than perfect and given that the percentage of 'occluded' eggs (described as 'opaque' eggs in Fletcher & Tregonning, 1992) is sometimes as high as 30% (Fletcher & Tregonning, 1992). This can be a major problem if they are subsequently miss-staged.

A range of interpretations of the initial drawings of Baker (1972) have been made during the staging of eggs for samples collected over the past 7 years, particularly for

relatively inexperienced sorters and especially for the 'occluded' stages. This has required many of the eggs being staged again. White and Fletcher (*in press*) produced a manual during this study which is designed to increase the speed, accuracy and uniformity in the staging of eggs by identifying problem areas and 'traps' for all personnel involved. Furthermore, frequent training is required if there is a relatively high rate of staff turnover. The manual should expedite such training and minimise the amount of re-staging that needs to be done.

Eggs from the Clupeidae are easily distinguished from other fish eggs, mainly by the large perivitelline space, the segmented yolk and having only a single oil globule. The main characteristics for live pilchard eggs (*Sardinops sagax*) are described below and are summarised from Baker (1972). If a full description of this species eggs is required the reader is referred to Baker (1972).

~ egg spherical

~ egg diameter between 1.32 and 1.70 mm (mean of 1.52 mm)

~ large perivitelline space (0.60 to 0.85 mm).

~ segmented yolk - thus the yolk has a rough, granular appearance

~ single oil globule - seen opposite the blastodermal cap (see egg description stage 3)

The egg descriptions offered in this report highlight the differences between preserved eggs and the live eggs described by Baker (1972). The descriptions of preserved eggs

are based on material which was placed in 5% buffered formalin and seawater for approximately 2 weeks and then transferred to 3% buffered formalin and seawater.

Eggs were considered *'occluded'* if the fluid surrounding the yolk and embryo had a white, slightly opaque appearance - in whole or in part. This is associated with various degrees of malformation or shrivelling of the yolk and\or embryo (or blastodermal cells). If the perivitelline space appeared slightly opaque but the yolk and embryo appeared normal and healthy, then the egg was not considered *'occluded'*.

The staging of *'occluded'* eggs is difficult but the same criteria apply in identifying the correct stage for *'occluded'* eggs as for normal, healthy eggs. In order to properly determine the stage for an *'occluded'* egg it may be necessary to extract the embryo from the egg case. The yolk of *'occluded'* eggs tends to disintegrate and/or dissipate, while the embryo tends to shrivel. Therefore it is often the case that an *'occluded'* egg has only a small amount of yolk and a malformed blastodermal cell mass or embryo. The degree of dissipation or shrivelling and malformation varies between eggs. It is best to stage *'occluded'* eggs based on the size and shape of the blastodermal cells or embryo and attempt to identify any of the characteristics found in normal, healthy eggs.

In order to properly establish the stage of the preserved and *'occluded'* eggs take note of the following points:

Stage 1

We are yet to find any stage 1 eggs in our samples. Therefore they are either difficult to identify or develop rapidly to stage 2.

Stage 2

The cells of the 8-cell morula are unmistakable in healthy eggs. In late stage 2 eggs (128 cell morula) the blastoderm is bulbous and distinct from the yolk in colour and shape. The main differences between this stage and stage 3 is that the blastodermal cells are larger and the blastoderm, as a whole, is more bulbous. Not many stage 2 eggs have been captured largely because little sampling was completed at night when they are most likely to be found.

Stage 3

The yolk of preserved specimens is not round but is a more elongate, oval shape. Stage 3 eggs are similar in appearance to stage 2 eggs in having a distinct, bulbous-shaped blastoderm, however, the blastodermal cells are not as large in stage 3 as in stage 2. In *'occluded'* eggs the yolk can deteriorate completely leaving only the blastodermal cells and the oil globule.

Stage 4

In preserved specimens shrinkage tends to make the blastodermal cells cover up to half or two-thirds of the yolk. The germ ring also becomes more evident and is seen as a pronounced ridge between the blastodermal cells and the yolk. A kidney-shaped, transparent area is seen within the blastodermal cells but on the opposite side to the development of embryonic shield. In *'occluded'* eggs, the yolk can deteriorate

completely leaving only shrivelled blastodermal cells or, if deterioration is not so advanced, then the blastodermal cells with a much reduced yolk is seen.

Stage 5a

Stage 5a eggs are easily misinterpreted since the embryonic shield is difficult to see in the early phase. The eggs may appear entirely lacking in structure apart from the yolk and a large perivitelline space. The egg must be turned on its 'side' to see the embryonic shield. It appears as a zone on the margin of the yolk, slightly more opaque than the rest of the yolk. This zone covers about one-third the diameter of the yolk. An area of transparency is also visible in stage 5 eggs opposite embryo development, however this is reduced in size compared to stage 4 eggs. The presence of the germ ring helps indicate an early stage 5 egg.

Stage 5b

In stage 5b eggs the entire embryonic shield is visible. The anterior region of the embryonic shield is distinct in colour and can be seen to rise above the surface of the yolk. The middle and posterior regions of the embryonic shield, however, remain flush with, and a similar colour to, the yolk. The blastopore closes so the germ ring is no longer evident. The main feature to note to separate this stage from stage 6 is the absence of the optic vesicles. This stage most likely corresponds to an early phase of stage 6 in Baker (1972). In a badly shrivelled, *'occluded'* egg it is possible to mistake a stage 5b egg for a *'occluded'* egg of higher development. This is because the malformation of the blastodermal cells and reduction in yolk tend to make the embryonic shield more obvious and to give it more shape. It is often easier and more

accurate to dissect out the yolk and embryo to properly establish the degree of embryo development.

Stage 6

The presence of the optic vesicles is the best criterion to establish stage 6 eggs but these are difficult to see in the early phase. They appear as oval shapes on either side of the head and are best viewed from above. Somites are visible in stage 6 preserved eggs. *'occluded'* eggs can, again, be easily misinterpreted as being a stage of higher development and extraction of the yolk and embryo from the egg case is advised.

Stage 7

The embryo is a distinct colour from the yolk and protrudes from the surface of the yolk for the entire length of the embryo. The tail does not separate from the yolk at any point. The end of the tail can reach the oil globule. The separation of the tail from the yolk is the best means of establishing whether an egg is stage 7 or stage 8. This is the case for both normal and *'occluded'* eggs.

Stage 8

The main feature to note is whether the tail has separated from the yolk. This may be seen by only a slight gap which appears just underneath the bulge in the tail.

The best way to determine the development of *'occluded'* eggs which are stage 8 or higher is to use the criteria outlined for normal-type eggs. It is important to note however that the yolk of *'occluded'* eggs is in a stage of deterioration and therefore the length of the tail may be deceiving.

Stage 9

The tail is separated from the yolk by one-third of the length of the embryo. However, the tail is not bent at an angle away from the dorso-ventral mid-line.

Stage 10

The tail of the developing larvae is bent away from the dorso-ventral mid-line at an angle of 45°.

Stage 11

The tail has grown to the extent where the tip may be parallel with the level of the hind-brain. The tip may curve in toward, but does not cross-over, the hind-brain.

Stage 12

The tip of the tail is curved in toward the head and crosses over the hind-brain. Specimens of this stage commonly have their tails preserved in positions other than the position it grows in the preceding stages. That is, the tail may not be parallel to the trunk and up toward the head, but may be in various other positions relative to the rest of the embryo. Presumably, this results from the increased ability of the embryo to move just prior to hatching.

2.5.2 Larval Staging

Larvae were also staged into three categories:

yolk-sac larvae	=	larvae with any remnant of yolk remaining.
PE larvae	=	no yolk is apparent and larvae are pre-flexion; no or little formation of dorsal fin.
PL Larvae	=	larvae are post-flexion; dorsal fin development obvious.

2.6 Standardisation of Data

The data obtained were converted to numbers per 200 m^3 of water using the flowmeter readings. However, due to occasional loss, damage or malfunction of flowmeters during some tows, readings were not obtained or considered erroneous. In these instances the flowmeter recording was estimated using a conversion factor. If a flowmeter reading was not obtained but the tow was considered to have no other problems, the average flowmeter reading for that cruise was used. ie.:

$$R_m = (\Sigma(R_i / L_i) / N) * L_m$$

where R_m = number of revolutions of flowmeter for record with missing value; R_i = number of revolutions of flowmeter for each record of the data set; L_i = length of tow for each record of the data set (i.e.: depth for vertical tows and duration for surface tows); N = number of records in dataset; L_m = length of tow for record with missing flowmeter value. A flowmeter reading was considered erroneously high or low if the number of flowmeter revolutions per length of tow unit occurred two standard deviations away from the mean for that data set. In this case the estimated flowmeter value for the conversion was taken back to one standard deviation away from the mean of that data set. Further problems encountered included that some samples were obtained from only one cod-end due to the contents of the other cod-end being lost. In these cases the calculated data for the remaining cod-end was doubled.

2.7 Other Work

Other research has also been undertaken using the samples collected, including a detailed examination of larval otoliths of pilchards and the identification and distribution of other fish larvae. This work was not part of the original project and has not been completed. Preliminary information is, however, provided in section 11.

Dates of Sampling	General Area Sampled	Net and Tow Type			Number of Stations conducted	Vessels Used
		Net Type	Mesh Size	Tow Profile		
July, 1992 (21-7-92 *to* 24-7-92)	Albany to Esperance	standard bongo	500	vertical	110	commercial vessels
January, 1993 (13-1-93 *to* 16-1-93)	Albany to Esperance	standard bongo	500	vertical	115	commercial vessels
July, 1993 (19-7-93 *to* 27-7-93)	Fremantle to Esperance	standard bongo	500	vertical	248	P.V. Baudin commercial vessels
January/February, 1994 (27-1-94 *to* 4-2-94)	Fremantle to Esperance	standard bongo	500	vertical	175	commercial vessels
July, 1994 (7-7-94 *to* 24-7-94)	Fremantle to Adelaide	1 metre square standard bongo standard bongo standard bongo standard bongo standard bongo EZ	1000 500 300 500 300 500 300	surface surface surface vertical vertical oblique layered	236	P.V. Baudin R.V. Franklin
December, 1994 (6-12-94 *to* 20-12-94)	Fremantle to Adelaide	0.5m square standard bongo standard bongo	1000 500 300	surface vertical vertical	57	R.V. Franklin
April/May, 1995 (26-4-95 *to* 7-5-95)	Esperance	CalVET standard bongo	300 500	vertical vertical	126	commercial vessels
July, 1995 (6-7-95 *to* 16-7-95)	Albany	CalVET standard bongo	300 500	vertical vertical	123	P.V. Baudin

Table 2.1: Details of plankton sampling surveys conducted between July 1992 and July 1995.

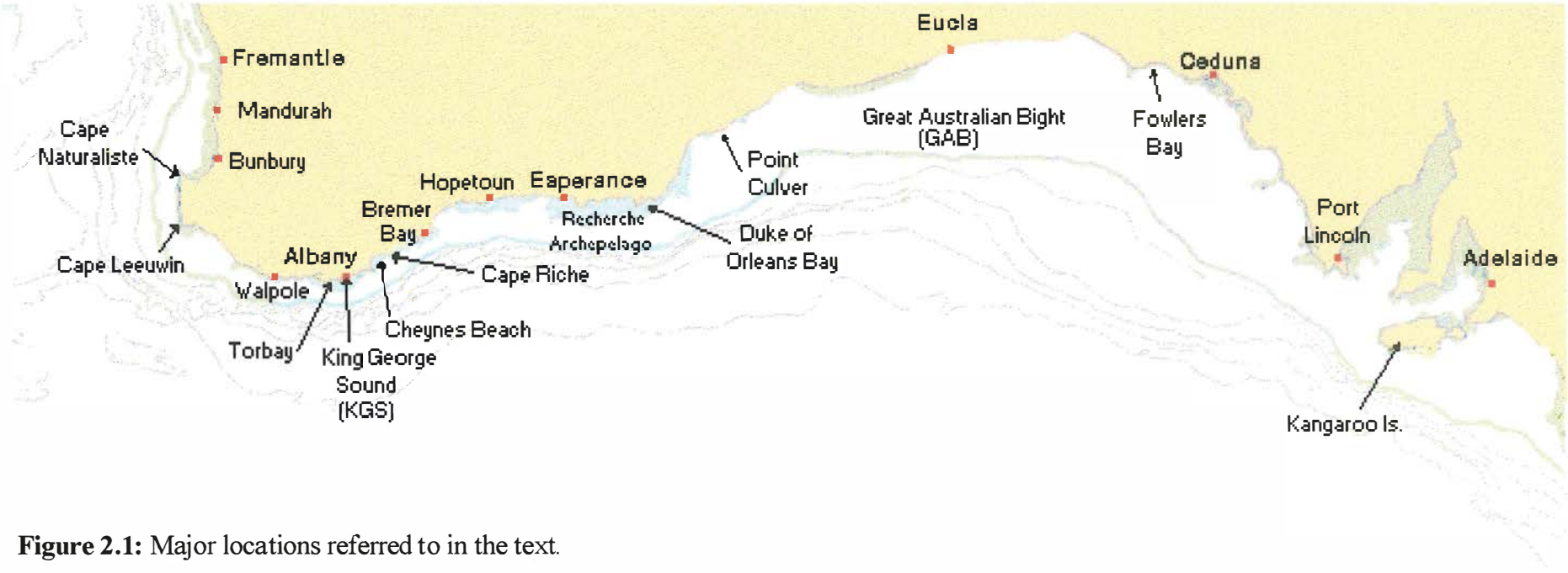


Figure 2.1: Major locations referred to in the text.

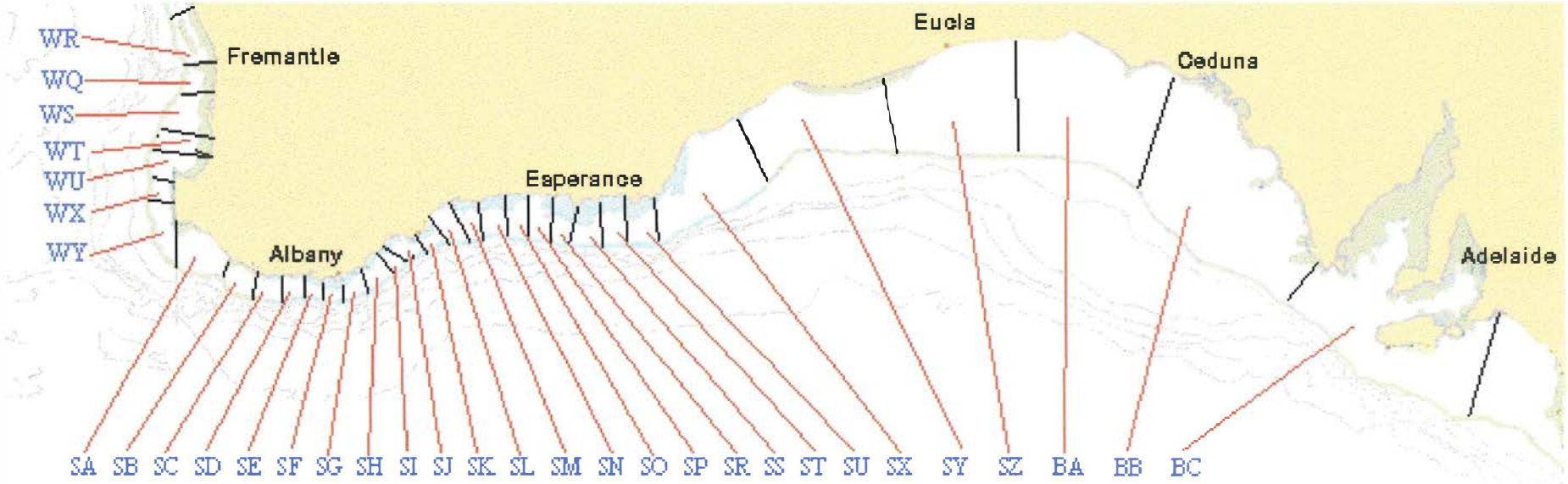


Figure 2.2: Boundaries of blocks into which sample stations were classified. Allocated block codes are shown in blue.

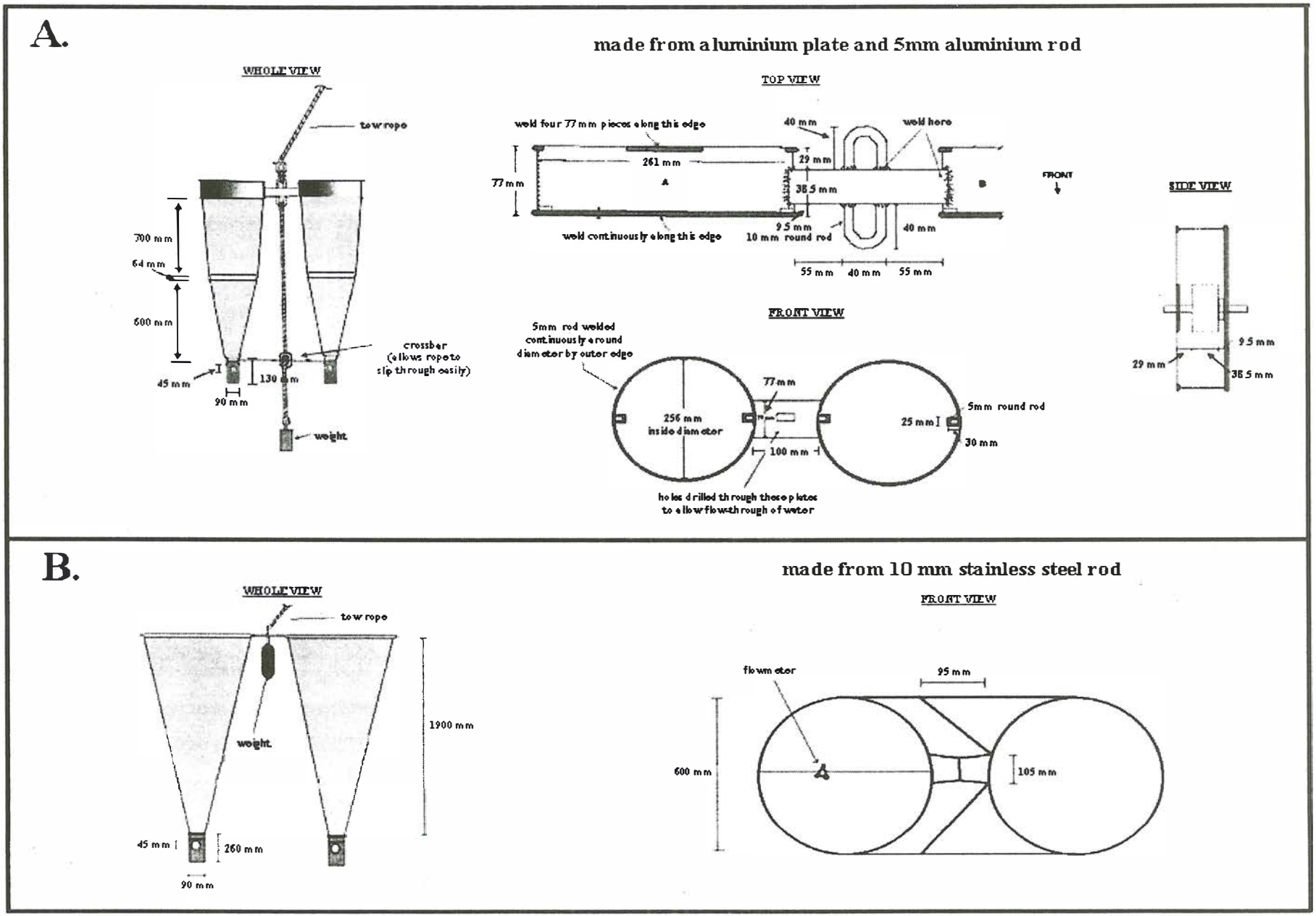


Figure 2.3: Details of Bongo net dimensions **A.** CalVET net [modified from Smith *et al.*(1985)] **B.** Standard Bongo net

3. Section 3 Pilchard Egg Development Rates

3.1 Introduction

A number of studies have examined the egg development rates of sardines, both *Sardinops* and *Sardina* (eg Sette, 1943; King, 1977). All were laboratory based with eggs cultured at different temperatures from which the relationships between the speed of development and temperature were developed. Our methods of sampling and on board facilities allowed egg culturing only once. However, the range of temperatures at which sardine eggs were found over the 6 seasons and the multiple locations sampled resulted in a large combination of temperatures and times of capture being recorded. These data were, therefore, examined to ascertain if any patterns could be discerned from which development rates could be determined or estimated. If patterns were found, these would be useful because they are based on data obtained under natural conditions rather than the potentially biased results obtained from eggs reared under laboratory conditions.

3.2 Methods

The data for all cruises were summed and divided into 6 groups based on sea surface temperature at the location of capture; < 16.5, 17.5, 18.5, 19.5, 20.5 and 21.5 °C. The abundance of eggs of each stage were plotted against the time of capture with the regression lines fitted using the median values of each stage to estimate the

development rates. The complicating factor was that sampling was generally restricted to the hours of 0600 to 1800 hr. Thus the night time period was only sampled during the cruise on the *RV Franklin* in July 1994. Furthermore a small culturing study was completed at approximately 18.0 °C from eggs collected at 0500 hr.

3.3 Results

3.3.1 Culturing

The eggs were caught at 0500 hr and were classified as early stage 3. They were maintained in small jars for the next 40 hours, with samples regularly removed and fixed in formalin. The last few eggs hatched at approximately 2300 hr the following day, approximately 48 hours after they were spawned (Fig. 3.1).

3.3.2 Plankton

There were clear patterns in the progression of stages with time (Figs. 3.2 to 3. 7). The earliest stages were seen just before or just after midnight (in the few samples that we have during this time period). The spread of times that each stage was present per day varied, but was generally between 4 and 8 hr. Some stages, however, appeared to be present through much of the day. Thus, for example, egg stage 8 at 18.0 °C was present in samples both early and late in the day. This was probably due to some having reached this stage late on their first day whilst others were still at this stage early their second day.

There was a clear increase in the speed of development with increasing temperature. Thus the times when the later egg stages were present varied greatly. These data are summarised in Fig. 3.8, whereby the regression of the median times for each stage were plotted for each temperature range (Table 3.1). The time for total development increased from 32 hours at temperatures > 20.5 °C to nearly 50 hours at temperatures less than 16 °C. This is similar to results obtained for *Sardinops sagax* in South Africa (Le Clus and Malan, 1995). The egg stages that should be present at each combination of time and temperature are located in Table 3.2.

3.4 Discussion

This study has confirmed that pilchards spawn at night, with the majority of spawning probably occurring before midnight. The development time of the eggs to hatching is dependent upon the temperature of the water in which they live. Thus, individuals in WA remain as eggs from 1.5 to 2 days. This difference may have implications for total rates of mortality and also for the calculation of egg production estimates, if sampling occurs on a 24 hr basis.

The summary table (Table 3.2) created to indicate the likely stages present, given the time of capture and the temperature of the water at capture, should assist greatly with the assignment of pilchard eggs to the correct stages during sorting. This is particularly for those that were 'occluded' (see section 2) at capture where the embryo has often begun to disintegrate and shrink. This latter category is often difficult to assess and in some samples they make up to 30% of the total eggs collected in a survey (Fletcher & Tregonning, 1992).

Temp C°	Intercept	Slope (hrs)
16.0	2.23	2.029
17.5	1.81	2.264
18.5	2.16	2.44
19.5	2.42	2.456
> 20.5	2.39	2.691

Table 3.1: Regression equations for the egg development rates.

Time (Hr)	< 16.0		17.0		18.0		19.0		20.0		21.0	
	Day-one	Day-two	Day-one	Day-two	Day-one	Day-two	Day-one	Day-two	Day-one	Day-two	Day-one	Day-two
200	2, 3	7, 12	2	7	2	8	2, 3	8	2	9	2	9, 10
400	2, 3	7, 8	2, 3	8	3	8, 9	2, 3	8, 9	2, 3	9, 10	2, 3	10
600	3	8 (9)	3, 4	8, 9	3, 4	9	3, 4	9, 10	3, 4	9, 10	3,4	10, 11
800	4	8, 9	3, 4	8, 9	4	9,10	4	9, 10	3, 4, 5	10,11	4, 5	10, 11, 12
1000	4	(8), 9	4	9, 10	4, 5	9, 10, 11	4, 5	10, 11	4, 5	10,11	4, 5, 6	11, 12
1200	5	9, 10	4, 5	9, 10	4, 5	10, 11	5, 6	11, 12	5, 6	11,12	5, 6	12
1400	5	9, 10	5, 6	10	5, 6	11, 12	5, 6	11, 12	6	12	6, 7, 8	**hatch**
1600	(5), 6	9, 10, 11	5, 6	10, 11	5, 6	11, 12	5, 6, 7	12	6, 7	**hatch**	7, 8	
1800	6	10, 11	6, 7	11	6, 7	12	6, 7	**hatch**	7, 8		7, 8	
2000	6, 7	10, 11	6, 7	11	6, 7	**hatch**	6, 7		7, 8		8, 9	
2200	7	11, 12	7	12	2, 7		2, 7		8		8, 9	
2400	2, 7	12	2, 7	**hatch**	2, 7, 8		2, 7, 8		8, 9		9	

Table 3.2: Likely pilchard eggs stages that will be present at each combination of time and temperature. If the eggs are staged outside of these criteria, check again. The stage in brackets is less likely to be the dominant stage.

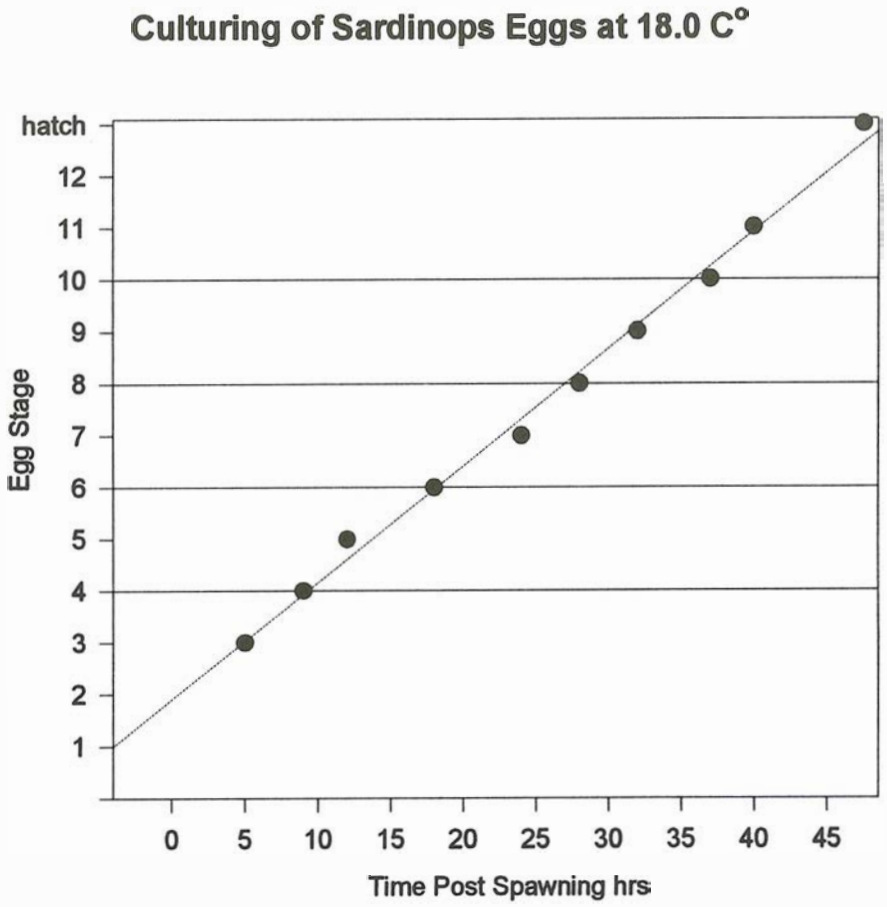


Figure 3.1: Development time for *Sardinops* eggs reared in the laboratory at approximately 18 °C. Spawning time was assumed to be 12 midnight.

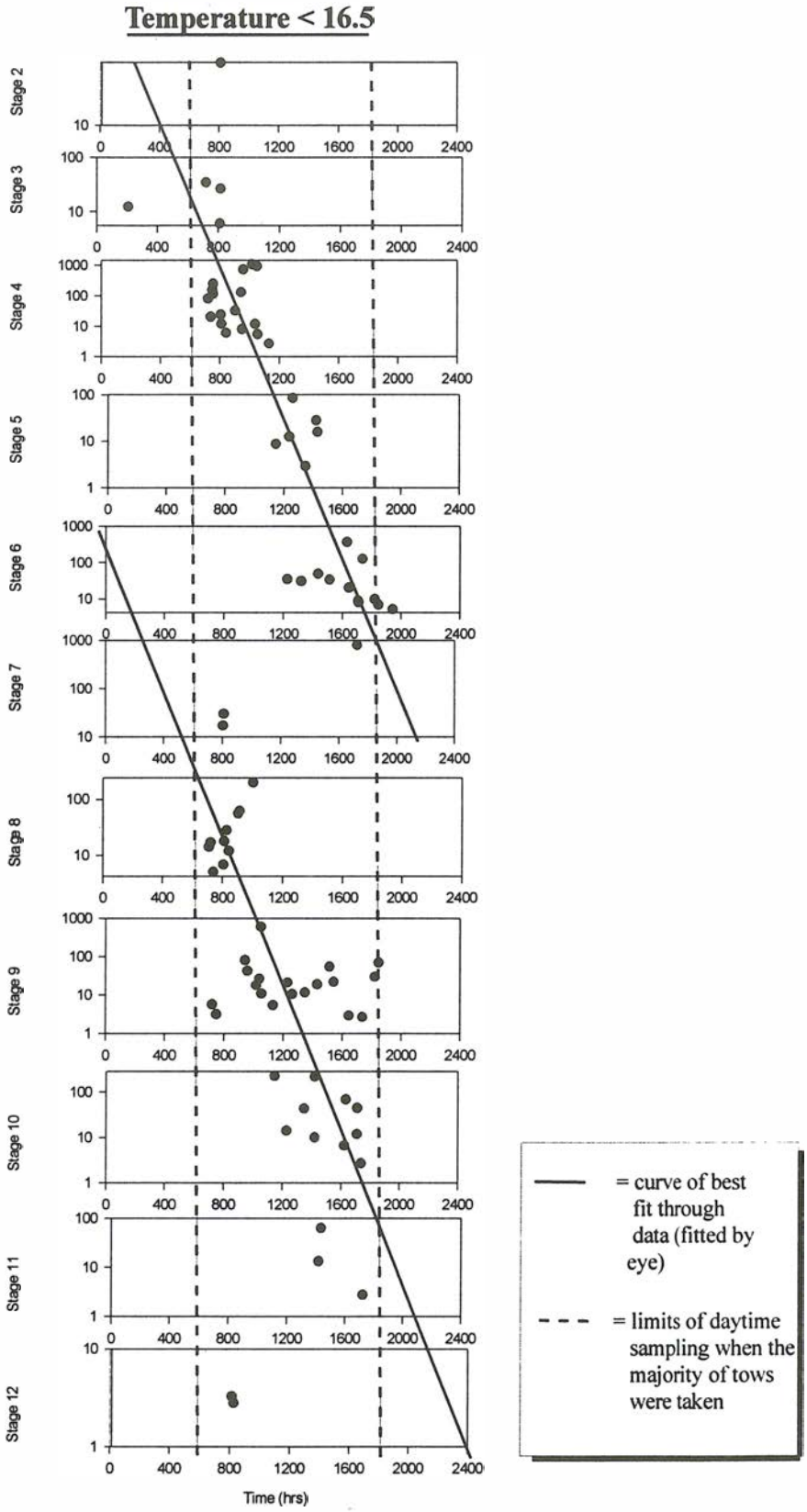


Figure 3.2: Abundance of each egg stage collected at temperatures below 16. 5°C plotted against the time of capture.

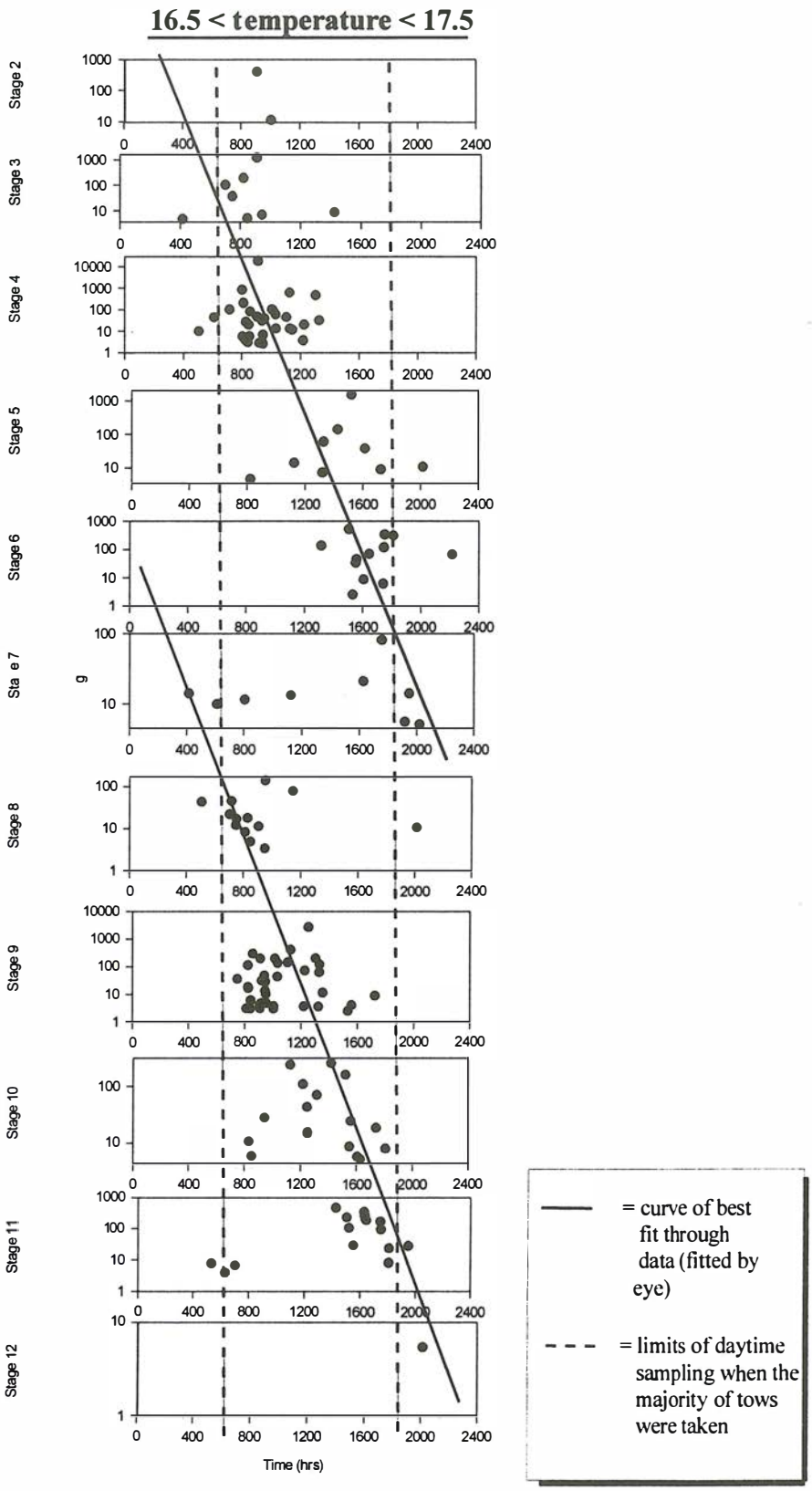


Figure 3.3: Abundance of each egg stage collected at temperatures between 16.5 and 17.5 C°plotted against the time of capture.

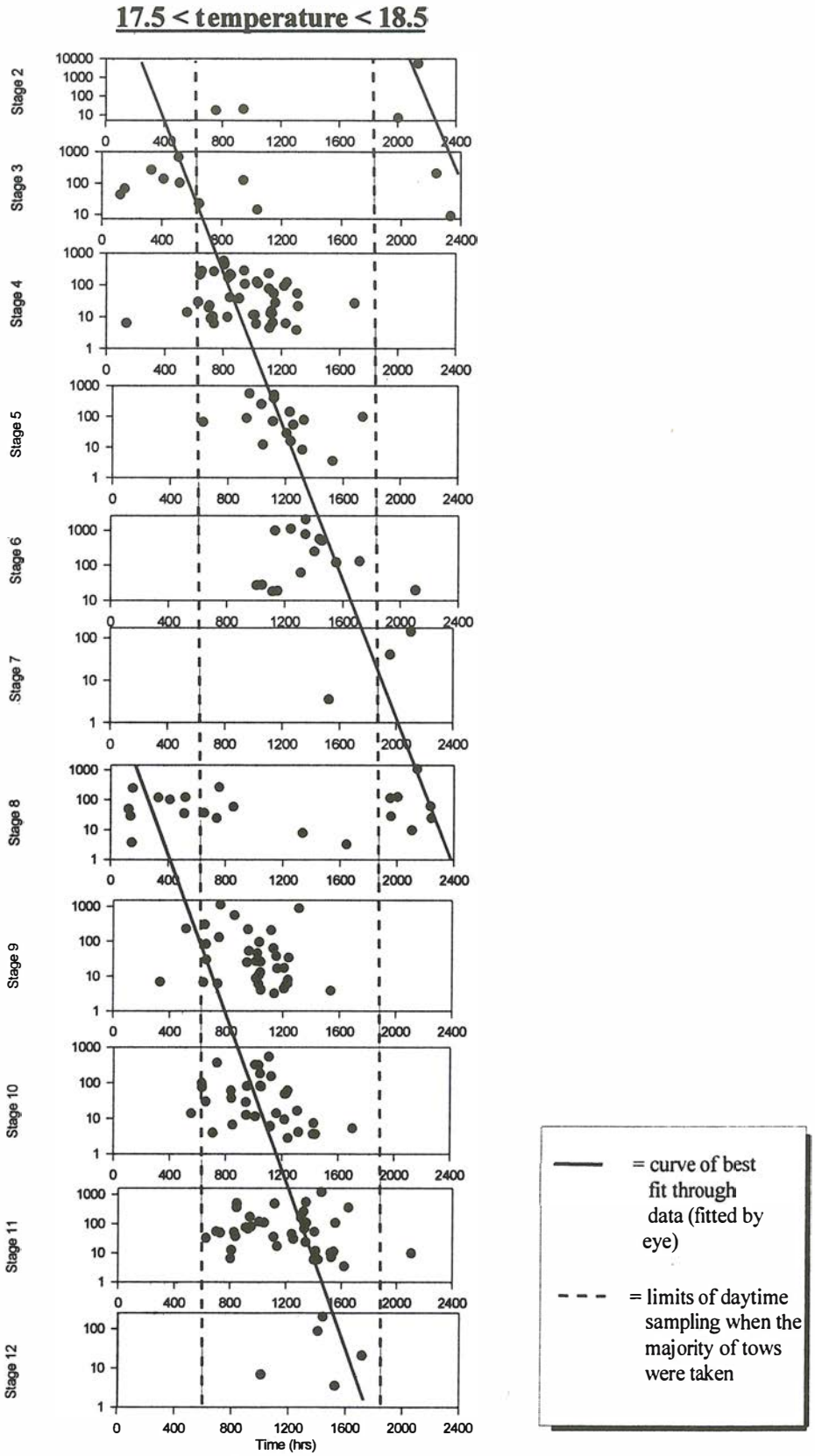


Figure 3.4: Abundance of each egg stage collected at temperatures between 17.5 and 18.5℃ plotted against the time of capture.

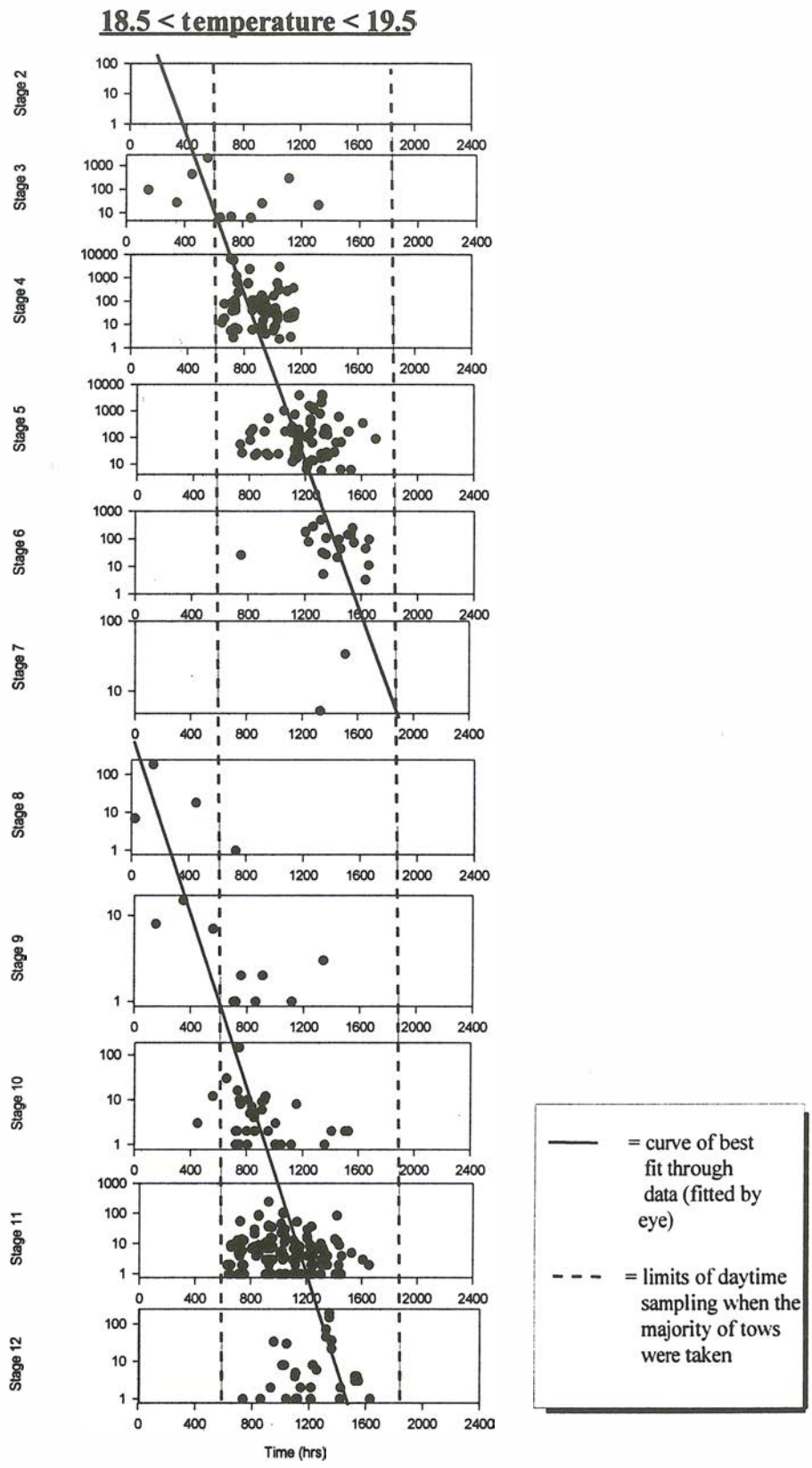


Figure 3.5: Abundance of each egg stage collected at temperatures between 18.5 and 19.5℃ plotted against the time of capture.

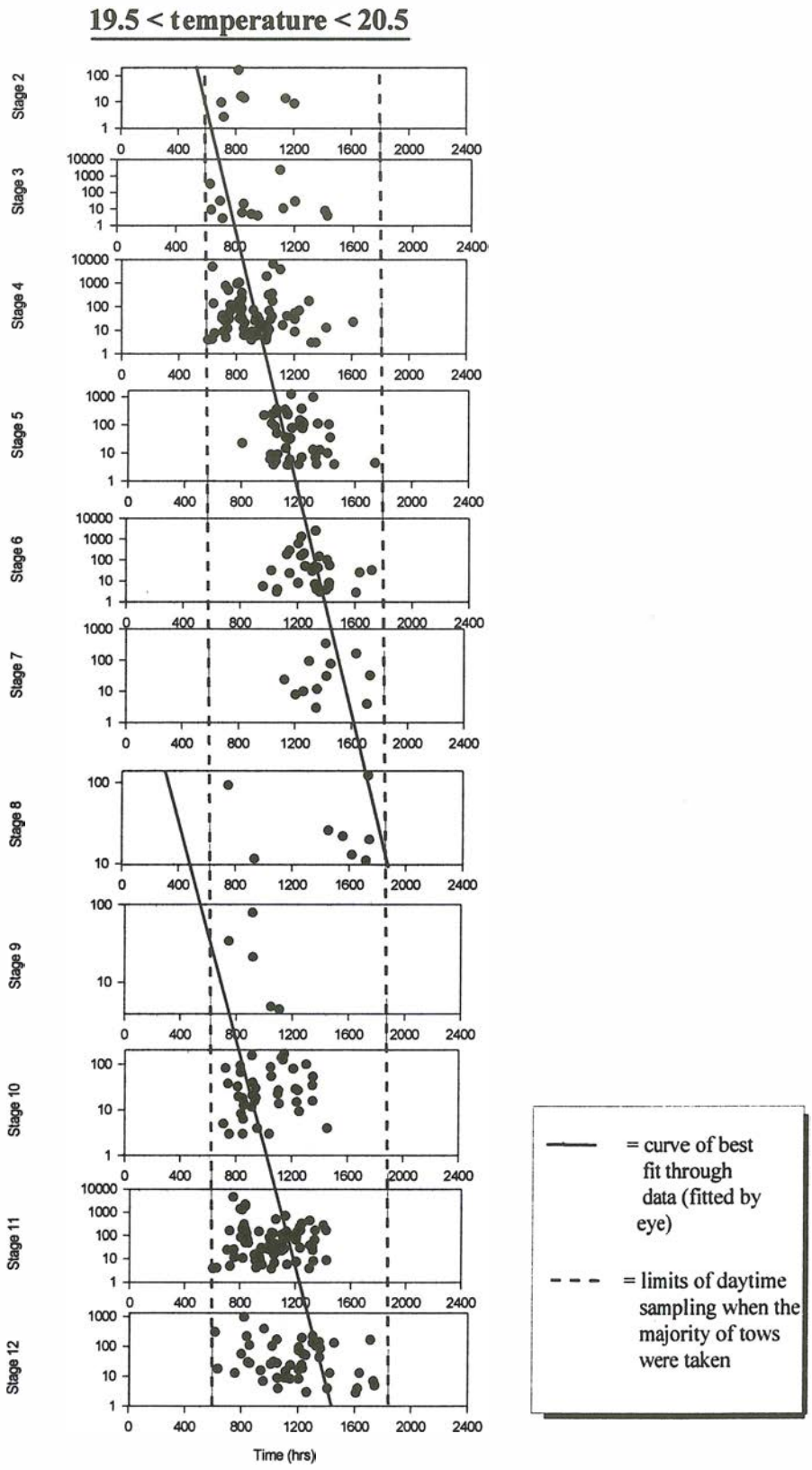


Figure 3.6: Abundance of each egg stage collected at temperatures between 19.5 and 20.5℃ plotted against the time of capture.

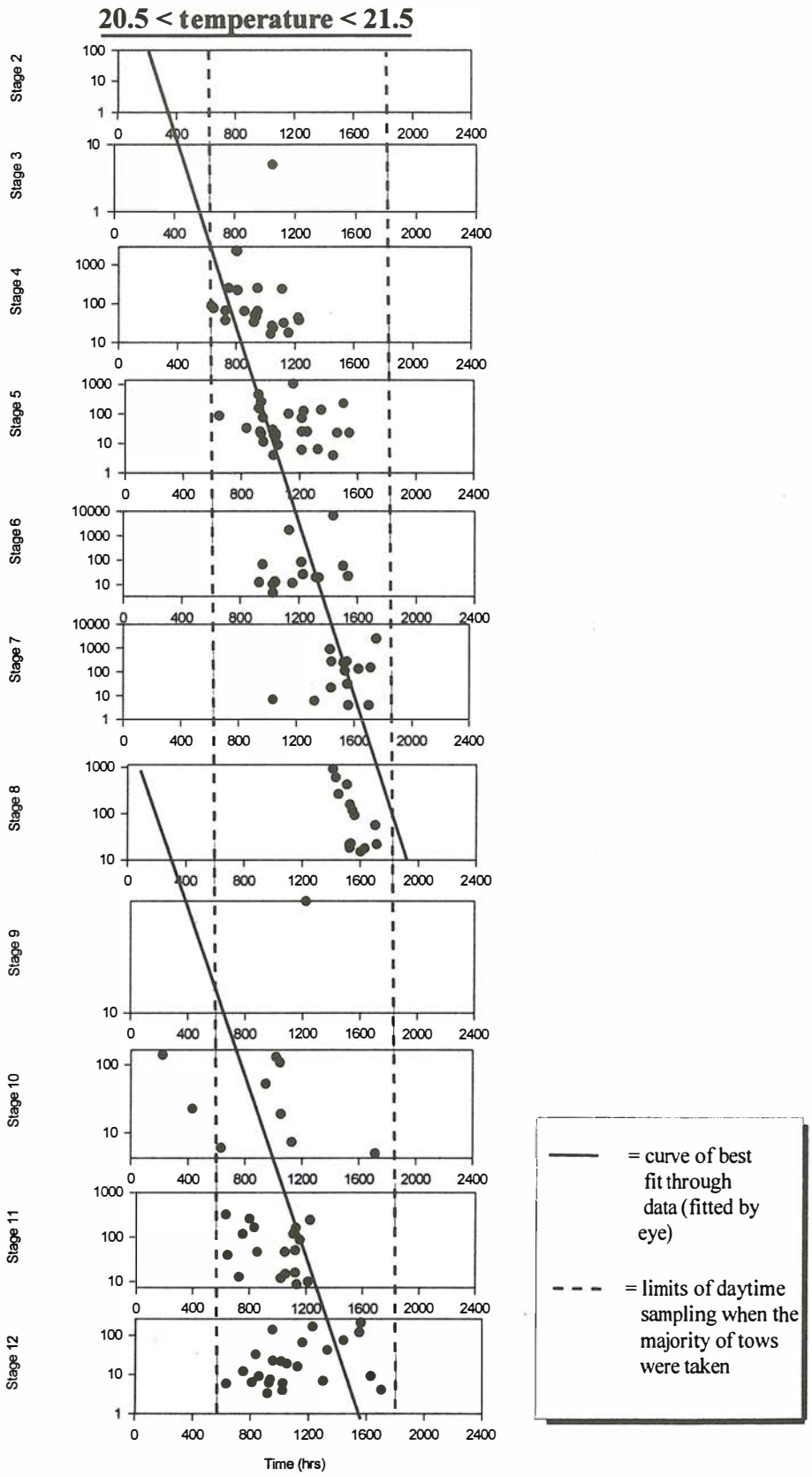


Figure 3.7: Abundance of each egg stage collected at temperatures between 20.5 and 21.5℃ plotted against the time of capture.

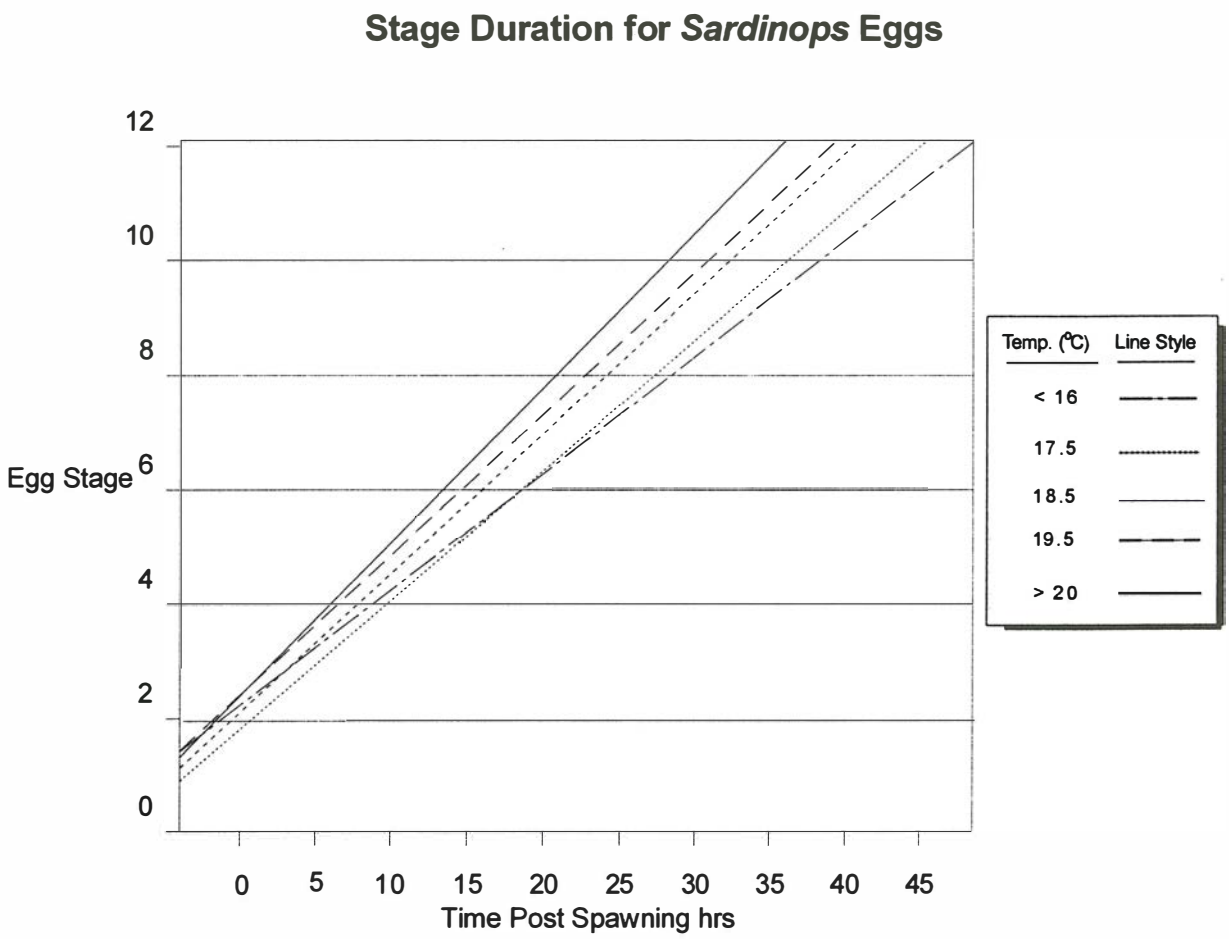


Figure 3.8: Development time for Sardinops eggs collected in 5 different temperatures ranges (from 16 - 21°C), estimated using the regression between the median value for each stage against hours post spawning. Spawning time was assumed to be 12 midnight.

4. Section 4: Spatial and Temporal Variations in the Distribution and Abundance of Pilchard Eggs and Larvae off Western Australia.

4.1 Introduction

In Western Australia, commercial fishing for pilchards is restricted to a number of relatively small areas compared to the total range of this species (Fletcher, 1990). With no access to a reliable research technique to capture adults (eg: mid-water trawling), an alternative strategy is required to obtain data on the distribution and abundance of pilchards in the extensive areas where samples could not be obtained from the fishery.

Indirect measures of fish abundance can be obtained using the distribution and abundance of eggs (see Hunter and Lo, 1993 for review). The use of plankton studies to examine stocks of pilchards is common, probably as a result of the widespread and highly mobile nature of these stocks (Heath, 1992). Consequently, the spatial, seasonal and yearly variations in the abundance of pilchard eggs and larvae off the south and south west coasts of WA were examined to provide information on the adult stock, including spatial structure and stock boundaries.

The spatial extent of pilchard spawning is related to the relative size of stocks (Wolf & Smith, 1985; Smith 1990). Thus, when the biomass of pilchards has been very large, spawning has extended well beyond the edge of the continental shelf (eg Watanabe *et al.*, 1996) and along a large proportion of the coastline (eg: Lluch-Belda *et al.*, 1989). By contrast, at low biomass levels, pilchard spawning is usually restricted to a number of small, coastal 'refuge' regions (eg Smith, 1990). The overall boundaries of spawning for WA pilchards should, therefore, provide some indication

of their stock size and the percentage of the stock(s) currently exposed to commercial exploitation.

In addition to determining the overall distribution of pilchards in WA, information on the pattern of spawning assists in the determination of separate pilchard stocks. Variations in the timing of spawning among areas and the degree of spawning segregation between regions can be used to indirectly assess the level of stock isolation (Heath, 1992). Differences have already been found in the cycles of gonad activity for WA pilchards (Fletcher, unpubl) which suggest that a number of separate spawning stocks may be present. Plankton surveys should provide detailed information on the regional and seasonal variations in the intensity of pilchard spawning along the coast from which any boundaries among spawning stocks may be discerned.

Plankton surveys should also enable detailed examinations of the early life history stages of pilchards. Oceanographic conditions within the area of spawning have often been hypothesised to have a large impact on the fate of planktonic stages and hence the numbers of juveniles that subsequently survive to recruitment. In particular, the abundance of many *Sardinops* populations have fluctuated greatly on a worldwide scale (Lluch-Belda *et al.*, 1989), usually associated with variations in the relative success of recruitment (Jacobson and MacCall, 1995). Information on the planktonic life history phases of pilchards could be used to investigate possible mechanisms to explain the impact of environmental fluctuations on the levels of recruitment.

Fletcher *et al.*(1994) have already shown that substantial advection of pilchard eggs and larvae is possible within the Albany region of WA when the Leeuwin Current is flowing strongly. It has also been hypothesised that the strength of the Leeuwin Current influencing this level of advection may subsequently affect the level of recruits that survive to enter the fishery (Caputi *et al.*, 1996). The present study will

examine whether a predictive relationship exists between the level of movement and the Leeuwin Current.

In this part of the study, we describe the plankton surveys which covered large regions of the south and south west coasts of Western Australia in both the winter and summer periods of 1992 to 1995. Whilst the planktonic stages of many species were collected, only pilchard material will be discussed in detail; the other species are presently being analysed with preliminary results in section 11.

4.2 General Methods

In total, 8 surveys were completed (see Table 2.1). These covered different lengths of the coast and in a few cases different towing methods and equipment were used. Thus, while the general methods for sampling were similar in all surveys (Section 2), specific differences within each survey are reported in the results section.

4.3 Results

The spatial distribution of the data of the planktonic groups are presented in a series of figures (Fig. 4.1 to 4.27). In these figures the value of any data at a particular geographical point is represented by a circle with a specific size and colour. The value to which this size and colour represent are shown in the legend associated with each picture. It is important to note the range of values represented in each legend to adequately make comparisons between surveys and\or groups. It is important to note in these figures also, that a small blue cross represents a zero value.

4.3.1 July 1992

Sampling Regime: A total of 110 stations were sampled between Denmark and Duke of Orleans Bay, using 4 vessels between 21-23 July 1992. In general, only the shelf region out to a distance of approximately 15 nm from the coast was surveyed because previous work in the Albany region had showed that this was the area where most pilchard eggs were located (Fletcher & Tregonning, 1992). In the area near Hopetoun, however, the inshore waters were considered too shallow (< 20 m) to conduct effective vertical tows. Therefore, the stations were moved further offshore to between 15 and 30 nm from the coast

General: Pilchard eggs and larvae were common throughout the survey area (Fig. 4.1a,b). Other species of larval fishes were present at all sampling locations but were most common near Albany and Hopetoun (Fig. 4.1c). During this survey, the Leeuwin Current appeared to be flowing with a strong jet of warm (19 °C) water located offshore of King George Sound (Fig. 4.1d, f). In the region west of Albany, the jet flowed close to the coast , but it was located out near the edge of the shelf east of Albany, with an offshoot south of Bremer Bay. Consequently, inshore regions east of Albany had relatively cool, 17.5°C water, especially near Esperance where these lower temperatures extended out 15 nm from the coast. Salinity measurements indicate that relatively low salinity (< 35‰) water was present in the region (Fig. 4.1e), consistent with a Leeuwin Current influence.

Day-one Pilchard Eggs: In the Albany region, concentrations of Day-one pilchard eggs were found inshore close to King George Sound and Cheynes Beach (ave. = 86 per $200m^3$; Fig. 4.2a). Few of this stage were located west of Torbay and few were found at the offshore stations. At Bremer Bay, large numbers of Day-one eggs were again found at inshore locations (ave.. = 111), but east, towards Hopetoun were located further offshore (ave. = 216). At Esperance, only a light scattering of eggs (ave. = 44) was found, however only the inshore part of the shelf was sampled.

Day-two Pilchard Eggs: The pattern for the offshore distribution of Day-two eggs was similar to Day-one eggs but their longshore distribution was consistent with them having been advected slightly to the east by the Leeuwin Current (Fig. 4.2b). Thus in the Albany region the main concentrations (ave. = 49) were near Cape Riche, with similar numbers found near Bremer Bay (ave. = 56). The largest concentration was at Hopetoun (ave. = 412) and relatively large numbers also occurred east of Esperance (ave. = 91; Fig. 4.3).

Pilchard Larvae: The distribution of all three stages combined showed evidence of having been moved consistently towards the east with a peak close to Bremer Bay, and a further peak past Hopetoun (Fig. 4.2b, 4.3).

4.3.2 January 1993

Sampling Regime: Due to inclement weather and a lack of vessels an abortive attempt to sample the region in the nominated time of December 1992 was subsequently postponed until January 13-16, 1993. Three boats were used to complete the 115 samples which were collected from Torbay (117 °E) to about Duke of Orleans Bay (122.5 °E). The Hopetoun region, however, was not sampled due to the breakdown of the vessel contracted to do this segment.

General: Pilchard material was collected in most areas but the majority was in the western part of the survey (Fig. 4.4a,b). Other fish larvae were common throughout the entire region, but the largest numbers were present offshore of Esperance (Fig. 4.4c). Satellite imagery indicated relatively uniform sea surface temperatures across the entire sampling area (Fig. 4.4f) with most temperatures in the range 20-21 °C. The slightly warmer waters found offshore of Esperance were associated with the larger numbers of larvae (Fig. 4.4b, c). Somewhat cooler temperatures were, however,

located in the area east of Esperance where little pilchard material was collected. Salinity samples were uniform across the entire sampled area except for a few inshore sites near Bremer Bay (Fig. 4.4e) with relatively high salinity water > 35.5‰.

Day-one Pilchard Eggs: Substantial numbers of Day-one pilchard eggs were located across the entire shelf region in the Albany area (ave. = 441; Fig. 4.5a); only in the furthest offshore samples (which were located over the edge of the shelf) were they absent. Day-one eggs were common in tows from Torbay across to Cape Riche (ave. = 470), but further east relatively few were collected (ave. < 150). In the Esperance region, this stage was mainly found on the outer half of the shelf with only a few collected at the inshore stations.

Day-two Pilchard Eggs: Substantially fewer of this stage were collected (ave. = 67; Fig. 4.5b). The peaks found in the Albany region were, however, located in approximately the same area or even slightly west of where the Day-one eggs were located (Fig. 4.6). Few Day-two eggs were found east of Cape Riche except for one sample close to the shelf break offshore of Esperance where the temperatures were slightly higher.

Pilchard Larvae: Unlike the eggs, there was a relatively even distribution of yolk sac larvae across the entire sampled region (Fig. 4.5c). The distribution of both post larval stages were, however, very similar to that of the eggs, with the main concentrations found in the western Albany region (Fig. 4.5c,d; Fig. 4.6).

4.3.3 July 1993

Sampling Regime: The area sampled during this survey was expanded to include the area between Esperance on the south coast and Fremantle on the west coast . This was

possible, largely through the use of the departmental patrol boat, *PV Baudin*, which sampled between Fremantle and Albany. A total of 248 tows were collected using 5 vessels over a period of 8 days (July 19-27 1993). The area near Hopetoun was again not sampled due to the breakdown of one of the survey vessels, and the inclement sea conditions in the region.

General: Pilchard eggs and larvae were common in only some areas of the locations sampled during this survey. The pattern for the other larval fish species was similar to pilchards with relatively lower numbers in the Cape Leeuwin to Albany region. The Leeuwin Current appeared not to be flowing strongly during the period of this survey (Fig. 4.7f). The satellite image shows that the main flow stopped just south of Cape Naturaliste (although much of the image is cloud affected). Sea surface temperature measurements taken on the vessels confirmed the higher water temperatures off the Fremantle region and relatively similar temperatures from Cape Naturaliste to Bremer Bay (Fig. 4.7d). Only the Esperance area appeared to have slightly cooler, more saline water (Fig. 4.7d, e). In general, salinity was relatively high (> 35.5‰) at all locations, particularly near Esperance.

Day-one Pilchard Eggs: Large numbers of Day-one pilchard eggs were collected on the west coast from Fremantle down to Cape Naturaliste (ave. = 570; Fig. 4.8a). These eggs were mostly located in offshore regions close to the shelf edge. Only small numbers were collected in samples taken close to the coast. Between Cape Naturaliste and Albany almost no eggs were found (ave. = 3; Fig. 4.9). Relatively large numbers were again collected in coastal regions between Albany (ave. = 100) and the area east of Bremer Bay (ave. = 171) with the biggest concentrations found near Cheynes Beach. Eggs were distributed extensively but less abundantly (ave. = 27) over the wide mid-shelf region (Fig. 4.8a) from east of Hopetoun to Esperance, however sampling was interrupted. East of Hopetoun, where sampling was interrupted, across

to Esperance, eggs whilst less abundant (ave. = 27), were distributed extensively over the wide mid-shelf region (Fig. 4.8a).

Day-two Pilchard Eggs: This stage had a very similar distribution to that of Day-one eggs with average densities off the west coast of 49 and off Albany of 145 (Fig. 4.8b; 4.9).

Pilchard Larvae: The abundance of the yolk-sac larval stage on the west coast was relatively low with evidence of small scale, southward advection (Fig. 4.8c, 4.9). Whilst this pattern was even more evident for early post larvae (Fig. 4.8d) there was little evidence that by the time they had become late stage larvae that large numbers had been transported to the south coast (Fig. 4.8e).

On the south coast, the majority of both yolk-sac and early post larvae were found in the Cape Riche region, close to where most of the eggs had been located (Fig. 4.9).

4.3.4 January 1994

Sampling Regime: The area between Esperance and Fremantle was again sampled with 175 samples collected using only two boats between 27 January and 4 February 1994. The majority of the work was completed from the *PV Baudin.*

General: Pilchard material was only common in the areas west of Bremer Bay, particularly off Albany and north of Cape Naturaliste (Fig. 4.10a, b). Similarly, other larval fish species were common from Fremantle to Cape Riche, but relatively few were found east of there (Fig. 4.10c). Satellite imagery indicated that the Leeuwin Current was not flowing (Fig. 4.10f). The SST measurements confirmed that a temperature of 21 °C was present across most of the survey area apart from local

heating at inshore locations on the west coast (Fig. 4.10d) and a cooler region offshore of Esperance. The salinity readings, however, indicate a more complex structure with values near 35.5‰ offshore of the west coast, but slightly higher salinities elsewhere (Fig. 4.10e).

Day-one Pilchard Eggs: On the west coast, Day-one pilchard eggs were common on the outer shelf from Fremantle to Cape Naturaliste (ave. = 91; Fig. 4.11a). A few were located near Cape Leeuwin but the next major concentration was near Albany. Day-one eggs were common out to the shelf break from Albany to Cape Riche (ave. = 162), but further east numbers were minimal (ave.=17; Fig. 4.12).

Day-two Pilchard Eggs: This stage was found off the Albany region (ave. = 102; Fig. 4.11b) but they were rare in all other locations, including the west coast (Fig. 4.12). On the south coast, the centre of their distribution was slightly west of where the concentration of Day-one eggs had been located (Fig. 4.12).

Pilchard Larvae: The concentrations of yolk sac larvae (Fig. 4.11c) and post larvae (Fig. 4.11d) on the west coast were largely restricted to mid-shelf and inshore regions. There was some evidence for transport of the stages south with the majority located south of Bunbury. This effect was most noticeable for the late stage post larvae, with most located between Cape Naturaliste and Cape Leeuwin.

On the south coast, the patterns of egg and larval abundance suggest a westward movement (Fig. 4.12). Thus, whilst the majority of eggs were found between Albany and Cheynes Beach, the main quantities of early and late stage larvae were located between Walpole and Cheynes Beach(Fig. 4.11c,d). Combined with the apparent southwards movement on the west coast, there was only a small break in the distribution of pilchard larvae of all stages between Fremantle and Cheynes Beach

(Fig. 4.10b). However, few pilchard larvae were found east of Cape Riche. (Fig. 4.10b).

4.3.5 July 1994

Sampling Regime: This survey sampled from Fremantle to Kangaroo Is. in South Australia. This was achieved using the *PV Baudin* from Fremantle to Albany and the *RV Franklin* from Adelaide to Albany. A total of 221 stations were sampled using a variety of towing methods and nets (see Table 2.1). Descriptions presented here for eggs and early stage larvae are based on vertical net hauls with the 300μm mesh, whilst decriptions for the late post larvae are based on 1000μm mesh surface tows. Additional data from the numbers of eggs collected in vertical tows completed in the Denmark to Bremer Bay region using the 500μm mesh nets were also used.

General Physical Data: The Leeuwin Current appeared to be only flowing moderately during this survey (Fig. 4.13). The sea surface temperatures (Fig. 4.13a) were relatively high between Fremantle and Albany. The temperatures were slightly lower east of Albany. The surface salinity readings suggest that water with lower salinity (< 35.5‰) was flowing off the shelf break on the west coast (Fig. 4.13b). On the south coast, however, inshore waters were relatively saline (> 36.0‰). The satellite image (Fig. 4.13c), and the ADCP measurements (Fig. 4.14) suggests that whilst the Leeuwin Current was present across the GAB, the usually strong signal and jets were not evident.

Day-one Pilchard Eggs: On the west coast, Day-one pilchard eggs were common across the entire shelf from Fremantle south to Cape Naturaliste (ave. = 329; Fig. 4.15a). Almost none were found in the region between Cape Naturaliste and Walpole (ave. = 4). Large concentrations were found again across the majority of the shelf

region from Albany to the end of the Recherche Archipelago, east of Esperance (ave. = 240). Across the GAB into SA, however, very few Day-one eggs were collected (ave. = 6).

Day-two Pilchard Eggs: The distribution of Day-two pilchard eggs was similar to Day-one except on the south coast there was evidence of some eastwards movement (Fig. 4.16). On the west coast there was little evidence of the eggs being advected south by the Leeuwin Current but they tended to be more inshore than the Day-one eggs (Fig. 4.15b).

Pilchard Larvae: Both yolk-sac and early post larvae of pilchards were abundant in the samples collected in the vertical tows completed on the west coast from Mandurah to south of Cape Naturaliste (Fig. 4.17a,b). Late stage post larvae were only common near Cape Naturaliste (Fig. 4.17c).

Few larvae were found between Cape Naturaliste and King George Sound. In vertical tows, yolk sac larvae were again common near Albany whilst post larvae were further east near Cape Riche and across to the end of the Recherche Archipelago. Only a scattering of larvae was collected from the vertical tows across the GAB. The surface net, however, collected a number of very large larvae off Albany and Esperance, and on the eastern side of the GAB offshore of Fowler's Bay (Fig. 4.18).

4.3.6 December 1994

Sampling Regime: Samples were collected using vertical tows with standard Bongo nets and surface tows with a 0.5 x 0.5m square net from Port Lincoln, South Australia to Fremantle, Western Australia between the 6-20th of December 1994 aboard the *RV Franklin*. Nineteen samples were completed across the GAB to Bremer Bay, all of

which were taken on the continental shelf. Fifteen samples were taken in an offshore loop to Walpole in oceanic waters. Subsequently, a number of across-shelf transects were completed with 25 tows taken between Walpole and Fremantle

General: Very little pilchard material was collected during this cruise (Fig. 4.20a). However, other fish larval species were common, particularly off the west coast (Fig. 4.20c). The sea surface temperature inshore on the west coast was 21.5 °C; this had declined to 17 °C in stations located on the shelf in SA (Fig. 4.19). The salinity readings, however, showed little difference across this distance with most readings above 35.5‰.

Pilchard eggs and larvae: The majority were collected on the west coast between Fremantle and Cape Naturaliste, particularly for the early larval stages. Few pilchard eggs of any age were collected, with only a few positive stations for either Day-one or Day-two eggs near Ceduna, Bremer Bay and Fremantle (Fig. 4.21, 4.22 a,b).

4.3.7 May 1995

Methods: This survey was completed in the Esperance and Albany regions during the period when the pilchard mortalities were occurring (Fletcher *et al.*, 1996). Sampling was completed from the 26 April to 7 May 1995 using 3 boats, 126 CalVET samples were collected, 105 in the Esperance region and 21 off Albany.

General: Pilchard eggs and larvae were found throughout the survey area, as were other fish larvae (Fig. 4.23 a,b,c). The pattern of sea surface temperatures, as indicated by the SST images, shows a fairly uniform temperature of 19 - 20 °C across the region. The actual temperatures measured during the survey showed this slight gradient more clearly with 18 °C inshore increasing to 20 °C near the shelf break.

Day-one pilchard eggs: These were found over most of the Esperance region from inshore regions out to the limit of sampling at the shelf break (Fig. 4.24a). There was no one specific region where eggs were significantly more abundant than others, and there was no relationship between egg density and the numbers of dead or dying pilchards (Fletcher *et al.*, 1996). The density of eggs collected was larger than had been found in this region on previous surveys with an average concentration of 254. The numbers found in the Albany region at this time were minimal (11).

Day-two Pilchard eggs: The distribution of Day-two eggs had a similar pattern to that seen for Day-one eggs (Fig. 4.24b). The average density of this stage in Esperance was 102 compared to 1 for Albany.

Pilchard larvae: All stages of pilchard larvae were common throughout the Esperance region with the main concentrations in the western area, particularly in more coastal locations (Fig. 4.24 c,d,e). The mean densities of the different categories were 44 (yolk sac), 134 (early post larvae) and 34 (late post larvae).

4.3.8 July 1995

Sampling Regime: The final sampling cruise for this project was completed in the Albany region between 6 and 12 July 1995. A total of 122 CalVET samples were collected from 116.5 to 118.5 °E. The region sampled extended from the coast to over the edge of the shelf.

General: Pilchard eggs and larvae were common at inshore locations in the Albany region (Fig. 4.25 a,b) with other fish larvae having a more patchy distribution (Fig. 4.25 c). A relatively strong Leeuwin Current was evident from the SST images taken

during this period (Fig 4.25f). Water temperatures were in the vicinity of 19 to 20 °C at the mid-shelf off Albany (Fig. 4.25d) with slightly cooler water elsewhere. Salinity measurements, however, suggest that the water was saline with all readings above 36‰.

Large numbers of Day-one pilchard eggs were found off Albany, with the majority mid-shelf to inshore (ave. = 215; Fig. 4.26a). None were found over the edge of the shelf. Day-two eggs were also relatively abundant (ave. = 104) but they appeared to have been advected approximately 20 nm to the east of where the Day-one eggs were located (Fig. 4.26 b). Few larvae were collected in the region immediately offshore of KGS. Most of the yolk sac larvae were abundant to the west of the Day-one eggs (Fig. 4.26 c). Post larvae found further away at approximately 60 nm from where the majority of Day-one eggs were located but because the sampling stopped, the true concentration may have been even further east (Fig. 4.26d). The distribution of these two larval stages was consistent with them having been pushed up against and along the coast.

4.4 Advection

To investigate the level of advection of eggs and larvae, the distribution of all pilchard stages in the Albany region were examined more closely. The surveys conducted during winter were examined separately as the Leeuwin Current is usually flowing much stronger over this period.

Winter: For the July 1992 survey, the pattern for the change in position in the main concentration of each stage supports the theory of a total easterly advection of 160 km (Fig. 4.27a). In July 1993, however, there appeared to be little trend, with the areas where the later stages were most abundant being both west and east from where Day-

one eggs were found (Fig. 4.27 b). In July 1994, an easterly movement of stages was again apparent, particularly for Day-two eggs and later with a total movement of 80 km (Fig. 4.27 c). Similarly, in July 1995, the pattern of movement was even stronger with peaks moving uniformly to the east from KGS to at least Cape Riche a distance of 100 km (Fig. 4.27 d).

Summer: The two detailed surveys conducted during summer, when the Leeuwin Current is not normally flowing, showed little easterly movement of the pilchard stages. This appeared to be the case in January 1993 (Fig. 4.27e), but some movement to the west occurred in January 1994.

The variations in the extent of this transport were examined in relation to the strength of the Leeuwin current, as measured by the Fremantle sea level (see Pearce, 1991 for justification) in June (Fig. 4.28). A clear and positive relationship was found between sea level height and the distance, in the eastwards direction, between where the majority of Day-one eggs were found and where the majority of post larvae were found ($r^2 = 0.88$, n = 7, $P < 0.01$).

4.5 Discussion

The combination of the large regions covered and the variety of temporal scales examined have provided a comprehensive account of the patterns of spawning and early life history of pilchards in WA. A number of consistent patterns in the timing and location of their planktonic stages were found, thereby addressing many of the main objectives of the study.

It has now been demonstrated that the distribution of pilchards in WA is consistently large, with most surveys finding recently spawned pilchard eggs over most of the

lower half of WA. Whilst an extensive distribution for pilchards had been suggested by previous studies (eg Blackburn, 1950), the present study has shown that during the one survey period, pilchard spawning can extend from the western end of the GAB to at least the Fremantle region on the west coast, a distance of 1300 km. Thus, pilchards are distributed over most of their range in WA and they appear to be present in all these regions during the entire year. Therefore, there is not one highly mobile stock which traverses the range on a cyclic fashion as found for the sardines off the west coast of America. In that regiom many of the adults migrated as far north as Canada during July to October and returned to southern California in winter to spawn during April-May (Clark, 1934; Janssen, 1938; Hart, 1943). Instead, the presence of large numbers of eggs off Albany and the west coast during summer shows that these stocks do not move to other locations at this time of the year when their catch rates are traditionally poor (Fletcher, 1991). Thus, the contention that the good catch rates in Esperance and the poor rates in Albany during summer were due to migrations of individuals between the two locations is not supported.

The longshore distribution of eggs, whilst extensive, was not uniform. The variations found did not appear to be random as there were areas on every survey where either discontinuities or concentrations of eggs were usually located. The largest, and most consistent gap was located between Cape Naturaliste, on the lower west coast, and Torbay on the south coast near Albany. In the five surveys which sampled all or part of this region, few eggs were collected. This pattern occurred in both winter and summer, but was especially noticeable in winter when almost no eggs were collected. This is strong evidence that there is a major discontinuity between the west and south coast pilchard.

Relatively large numbers of pilchard eggs were generally found offshore of KGS during all surveys. This pattern was also found during previous surveys of the region (Fletcher and Tregonning, 1992; Fletcher *et al.*, 1994) and supports the hypothesis

that this region tends to concentrate adult pilchards, particularly during winter (Fletcher, 1992). Inshore concentrations of eggs were also located in the region near Bremer Bay. In the Esperance region and off the west coast, however, pilchard eggs were usually more widely distributed, possibly a reflection of the increased shelf width.

A further discontinuity in the distribution of pilchard eggs was found during summer, whereby relatively few eggs were found east of Cape Riche. This supports the differences found in the pattern of gonad indices among regions with Albany and Fremantle having a bimodal cycle, suggestive of two spawning periods (summer and winter), whereas Bremer Bay and Esperance only have single cycles (autumn/winter only).

The sampling completed in May 1995 further clarified the temporal variation in spawning along the coast. Large numbers of eggs were found off Esperance, yet at Albany, almost none were found. Thus, the major time of spawning for Esperance appears to be May continuing through to July. At Bremer Bay, the major period is probably June -July. Whilst at Albany, the main spawning times are from June-July in winter and December to February in summer, with similar times for the west coast stocks.

The extensive sampling completed during July 1994, which traversed the entire GAB region, showed that the GAB does not form part of the WA spawning stock. Thus almost no pilchard eggs were collected east of Pt Culver with this pattern continuing across to where sampling ended near Kangaroo Is. This region can, however, contain large numbers of pilchard eggs during January-March (Blackburn, 1950; Hoedt, pers. comm.).

The extensive distribution of spawning along the WA coast is not matched in the offshore direction. Spawning was almost totally restricted to continental shelf regions. Numerous tows were completed off the edge of the shelf, with some 100 km beyond the shelf, but pilchard eggs were found rarely and then only in the samples close to the edge.

On the shelf, the location of the eggs varied with area and season. In the Albany area during winter, eggs were mostly on the inner part of the relatively narrow shelf, with few found more than 10 nm from the coast. This had been found in previous surveys in this region (Fletcher & Tregonning, 1992, Fletcher *et al.*, 1994). During summer in this region, the offshore extent of spawning appeared to be wider with eggs found out almost to the shelf edge. Such a seasonal increase in the spatial extent of the stock may provide the reason for the substantially lower catch rates seen at this time (Fletcher, 1991, 1992).

Spawning was more extensive off both the west coast and Esperance regions, where the shelf is wider. In contrast to Albany, the majority of spawning in these areas was found to be close to the edge of the shelf, with relatively little spawning in inshore waters. This has implications for the fisheries in these two areas, particularly for the west coast where fishing has until recently been restricted to coastal regions and there have been large fluctuations in the apparent availability of pilchards in the Fremantle area. With the introduction of new entrants and larger boats, fishing has moved offshore and catches have increased dramatically.

The fate of the pilchard eggs depended upon where and when they were spawned. On the west coast there was evidence of only minor southward transport despite the presence of the southwards flowing Leeuwin Current close to where the majority of the eggs were found. Much of the movement appeared to be inshore rather than

southwards. Thus, the expected large scale transport of pilchard material from the west to the south coast was not supported.

On the south coast, substantial eastwards transport of eggs and larvae occurred during winters where the Leeuwin Current was flowing strongly. This confirms the relationship found by Fletcher *et al.* (1994). By contrast, in years when the Leeuwin Current was weak (1993) or during the summer when it is not flowing, eastwards advection was minimal and in January 1994 there was evidence that movement to the west was occurring. This was probably a result of the persistent SE winds which occur during this period of the year.

The scale of transport on the south coast may be large. Between the first egg stage and early post larval phases individuals appeared to be moved up to 150 km. The extent of this advection appeared to be tightly related to the strength of the Leeuwin current with planktonic stages likely to be moved well beyond the apparent boundaries of the adult stocks that spawned them. Thus, the late stage pilchard larvae that were found at the eastern end of the GAB could have been transported to this location from the May spawning off the Esperance region. Counts of their daily rings were consistent with them being approximately 2 months old (see Section 11). Consequently there is likely to be substantial interchange between the stocks at this stage of their life history, even if there is separation during the adult phase. This mixing is consistent with the lack of distinct genetic differences among regions found by the electrophoretic study completed by Dixon *et al.* (1993).

The variation in the level of transport found between seasons and among years within seasons could play an important role in the subsequent survival of these stages and hence affect recruitment levels to the fishery in future years. Fletcher (1995) found that recruitment levels of 2 year old pilchards into the Albany and other south coast fisheries varied by an order of magnitude during the 1988 - 1995 period. There also

appeared to be variations in the timing of recruitment, possibly as a result of fluctuations in the relative success of the different spawning seasons. Caputi *et al.* (1996) provided a preliminary analysis of the relationship between the strength of the Leeuwin Current and the relative strength of recruitment which showed that strong LC years were associated with poor recruitment of juveniles two years later. However, more data are required to substantiate this relationship.

4.5.1 Implications for Fisheries Management

1. Pilchards have a widespread but patchy longshore distribution in WA which is restricted to the continental shelf waters. This pattern is consistent with only small to moderate stock sizes.
2. There is no evidence of mass migrations of individuals on a seasonal basis; areas where catch rates are seasonally low still have substantial numbers of eggs.
3. The west and south coast stocks of pilchards are largely separate spawning stocks with a consistent gap in spawning between Cape Naturaliste and Torbay, there is little transport of planktonic stages between the two regions.
4. During winter, the south coast has a number of areas where concentrations of eggs were consistently found, mainly off KGS and a further concentration near Bremer Bay.
5. During summer, spawning was largely restricted to the regions west of Cape Riche.
6. The main period of spawning off Esperance was shown to occur in May, which is earlier than the remainder of the south coast.
7. The WA spawning stock appears to end at the beginning of the GAB. Little spawning was found in the GAB region during July 1994.
8. Substantial transport of the planktonic stages of pilchards occurs during winter when the LC is flowing. Eggs spawned off KGS may be moved > 100 km by early

larval phases. There was some evidence that larvae found in the eastern GAB were possibly from spawning that occurred off Esperance 2 months earlier.

9. The extent of this easterly transport is related to LC strength which may in turn affect the subsequent level of recruitment of juvenile pilchards to the fishery two years later.

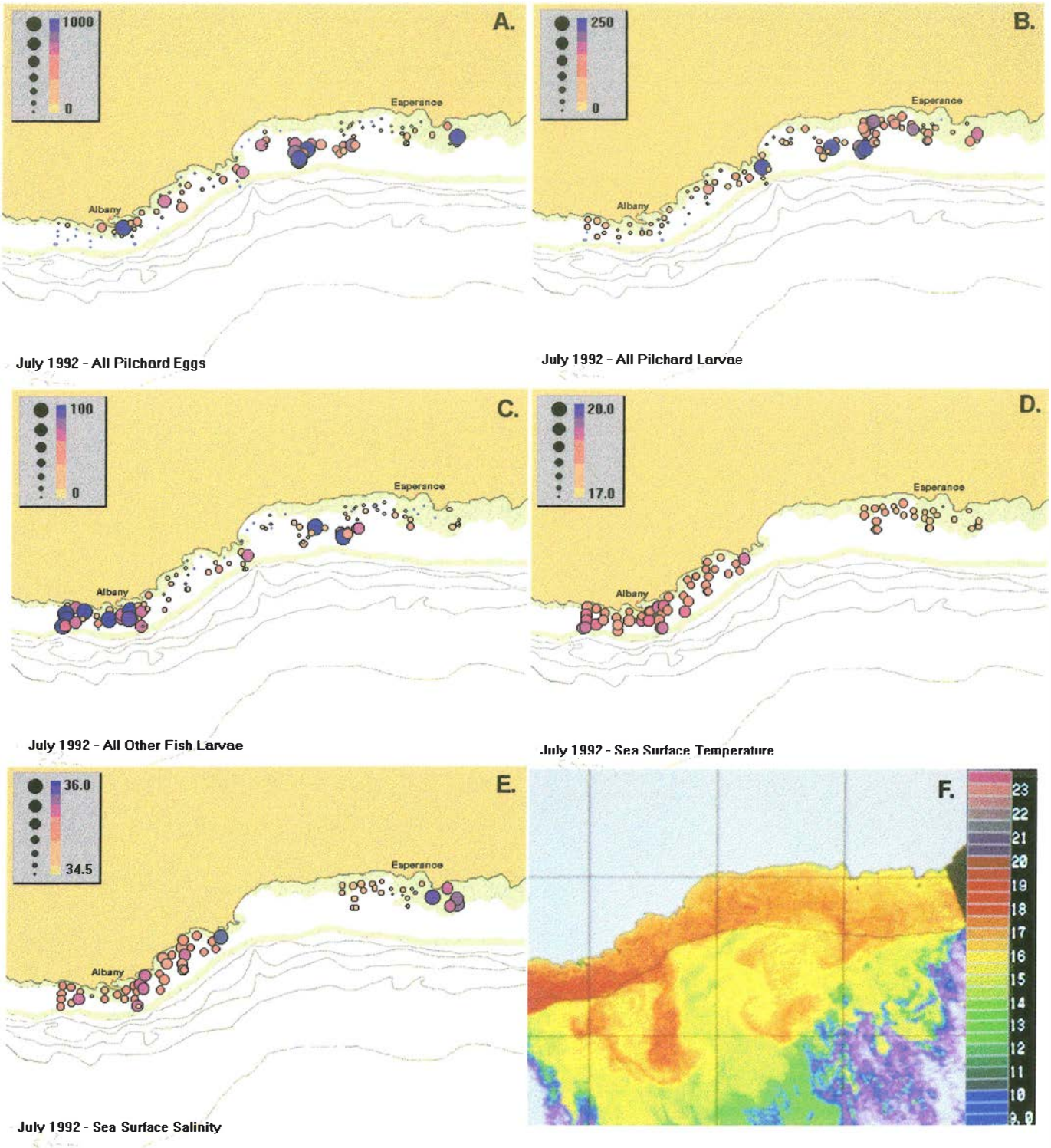


Figure 4.1: General plankton and physical data distributions during July 1992 field trip. **A.** *All pilchard eggs.* **B.** *All pilchard larvae.* **C.** *All other fish larvae.* **D.** *Sea surface temperature.* **E.** *Sea surface salinity.* **F.** *NOAA satellite image from 15/7/92 showing sea surface temperature.*

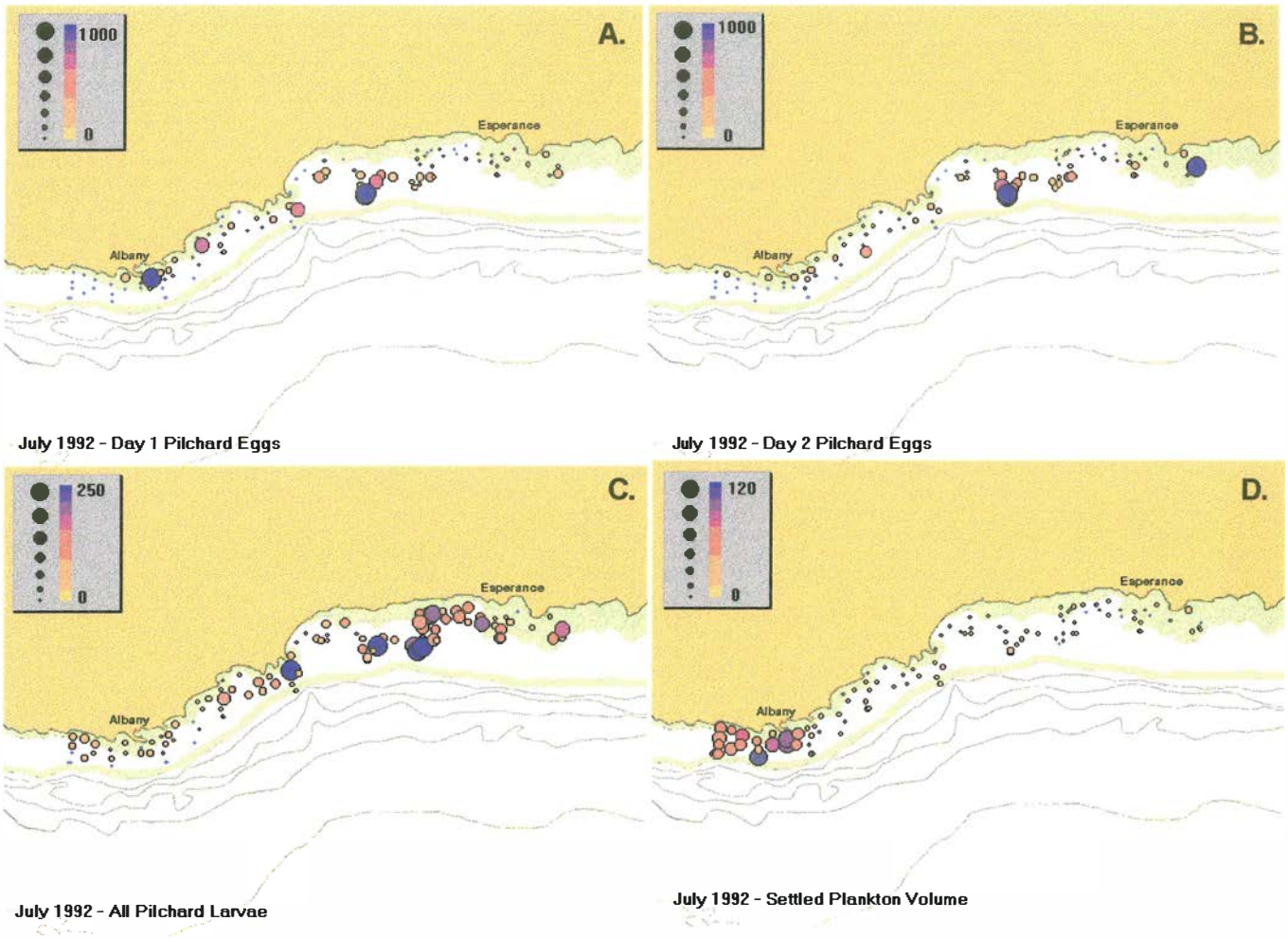


Figure 4.2: General plankton data distributions during July 1992.
A. *Day 1 pilchard eggs.* **B.** *Day 2 pilchard eggs.* **C.** All pilchard larvae **D.** *Settled plankton volume*

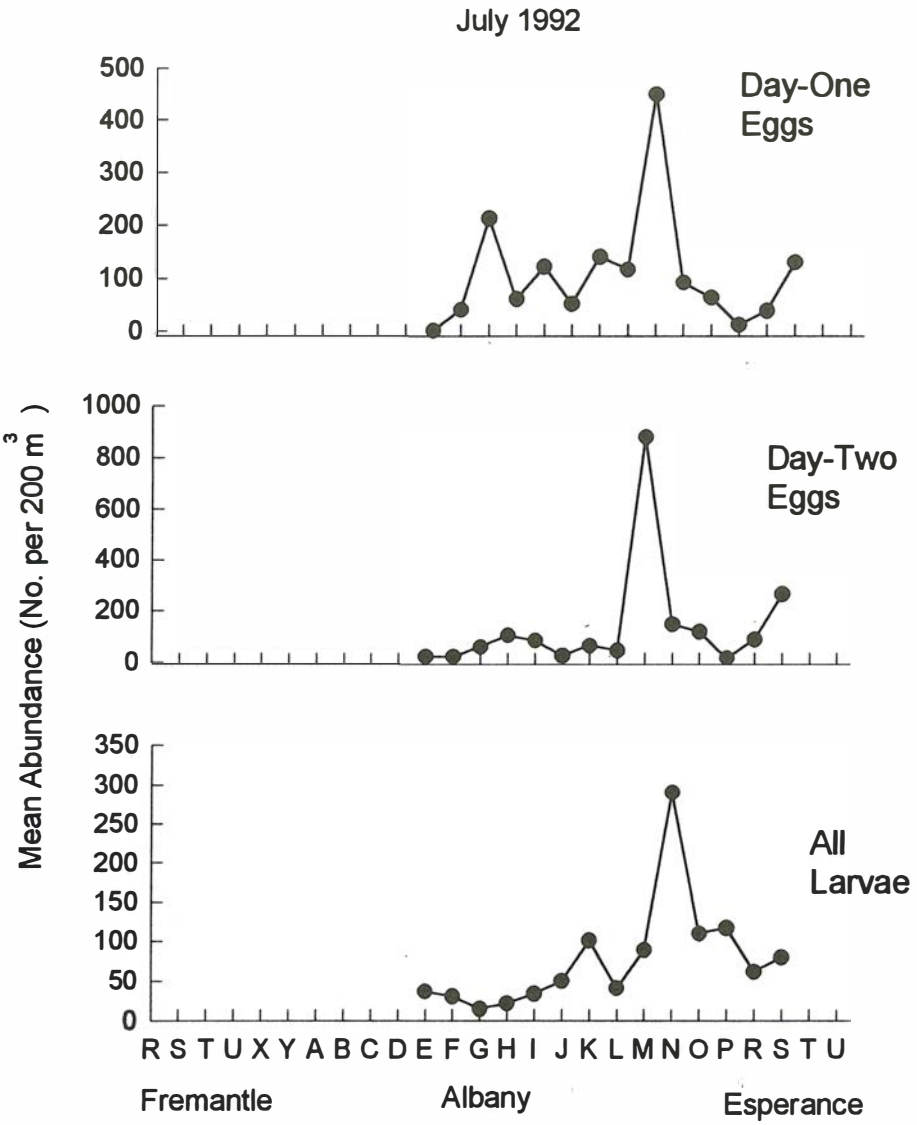


Figure 4.3: Relative distribution of pilchard stages Day-one, Day-two eggs, Yolk-sac and post larvae collected in the July 1992 survey. The blocks refer to the sampling blocks detailed in Fig. 2.1.

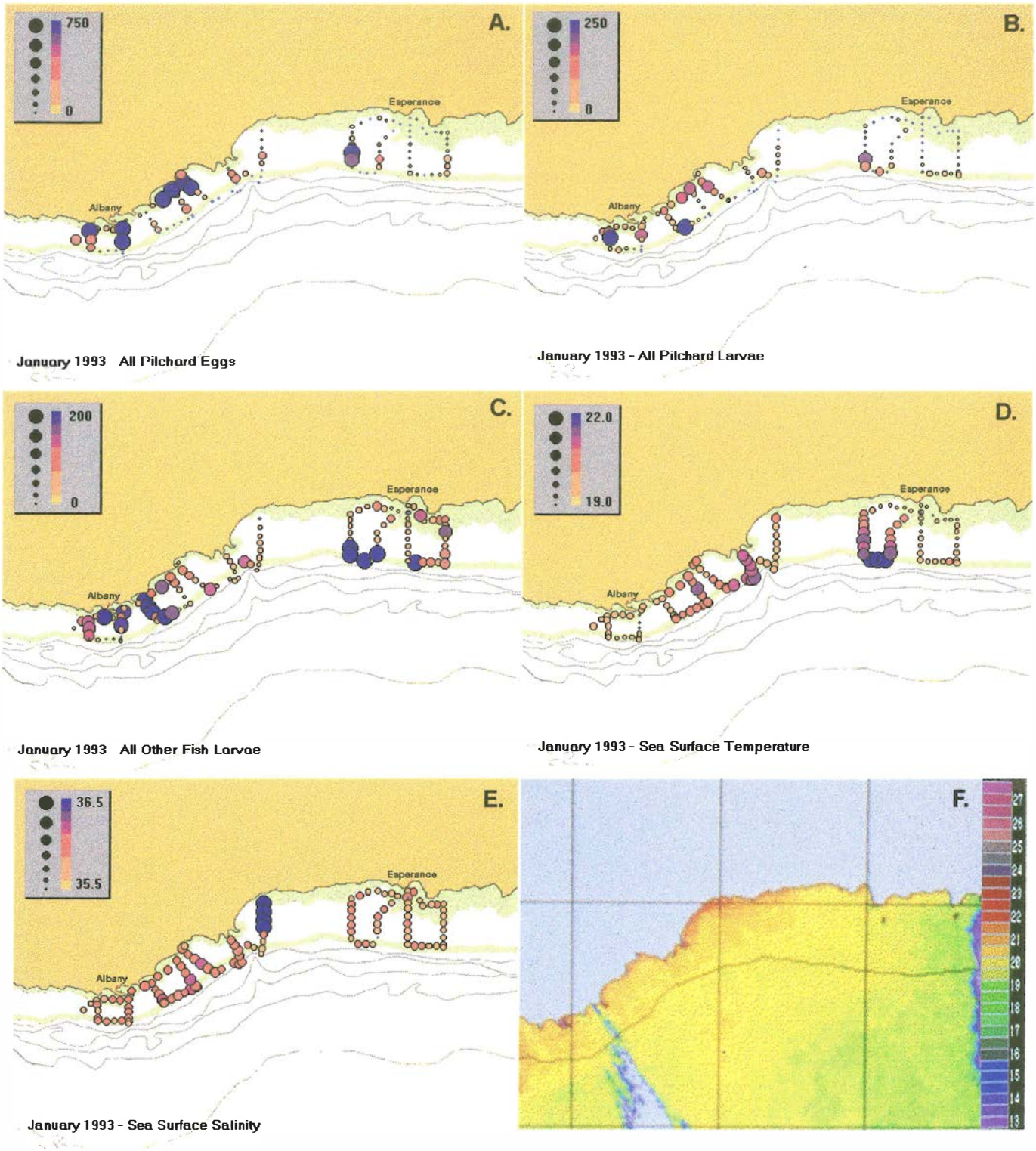


Figure 4.4: General plankton and physical data distributions during January 1993 field trip. **A.** *All pilchard eggs.* **B.** *All pilchard larvae.* **C.** *All other fish larvae.* **D.** *Sea surface temperature.* **E.** *Sea surface salinity.* **F.** *NOAA satellite image from 21/1/93 showing sea surface temperature.*

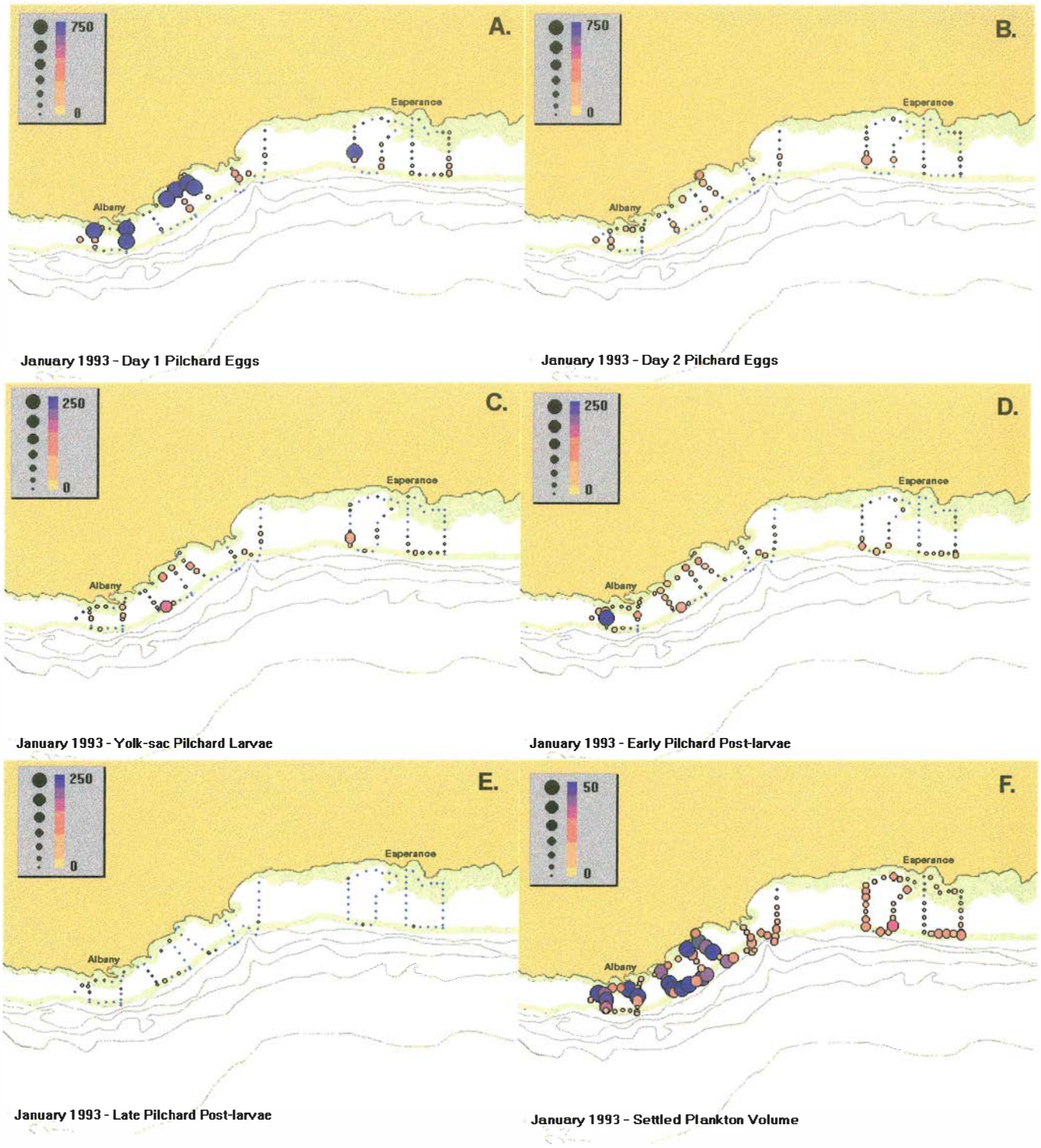


Figure 4.5: General plankton data distributions during January 1993 field trip.
A. *Day 1 pilchard eggs.* **B.** *Day 2 pilchard eggs.* **C.** *Yolk-sac pilchard larvae.*
D. *Early pilchard post-larvae.* **E.** *Late pilchard post-larvae.* **F.** *Settled plankton volume.*

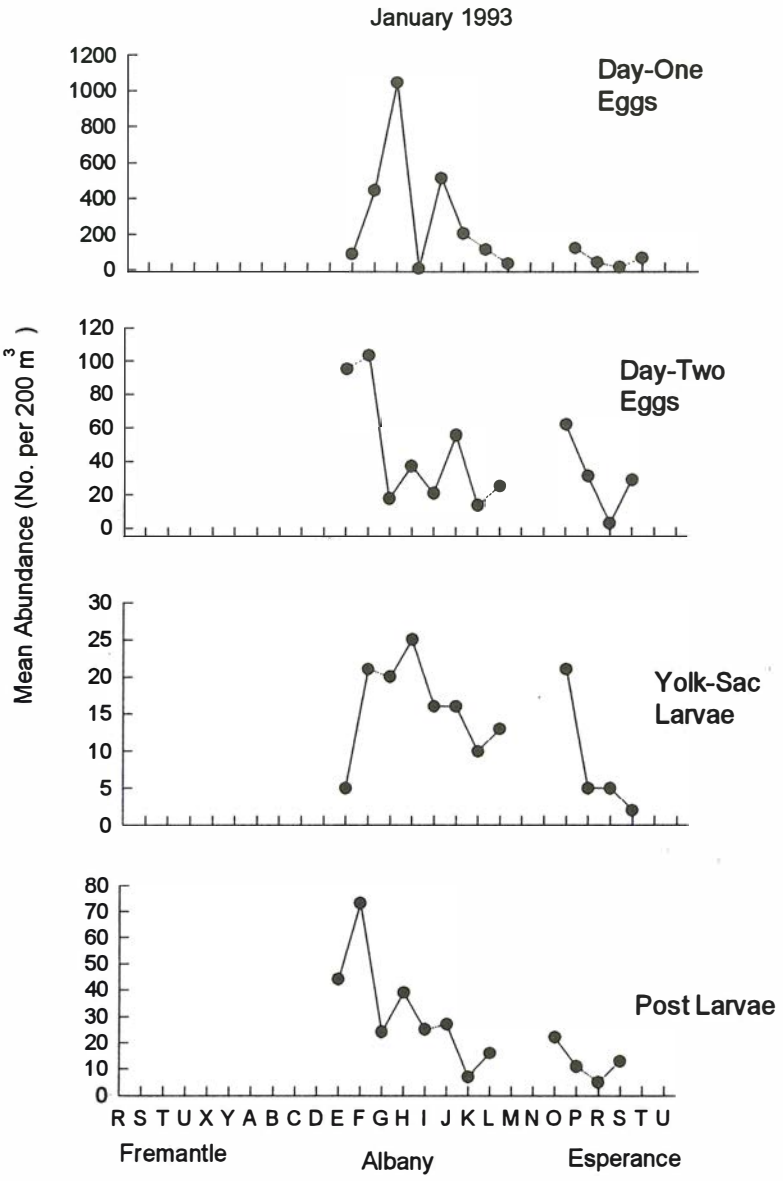


Figure 4.6: Relative distribution of pilchard stages Day-one, Day-two eggs, Yolk-sac and post larvae collected in the January 1993 survey. The blocks refer to the sampling blocks detailed in Fig. 2.1.

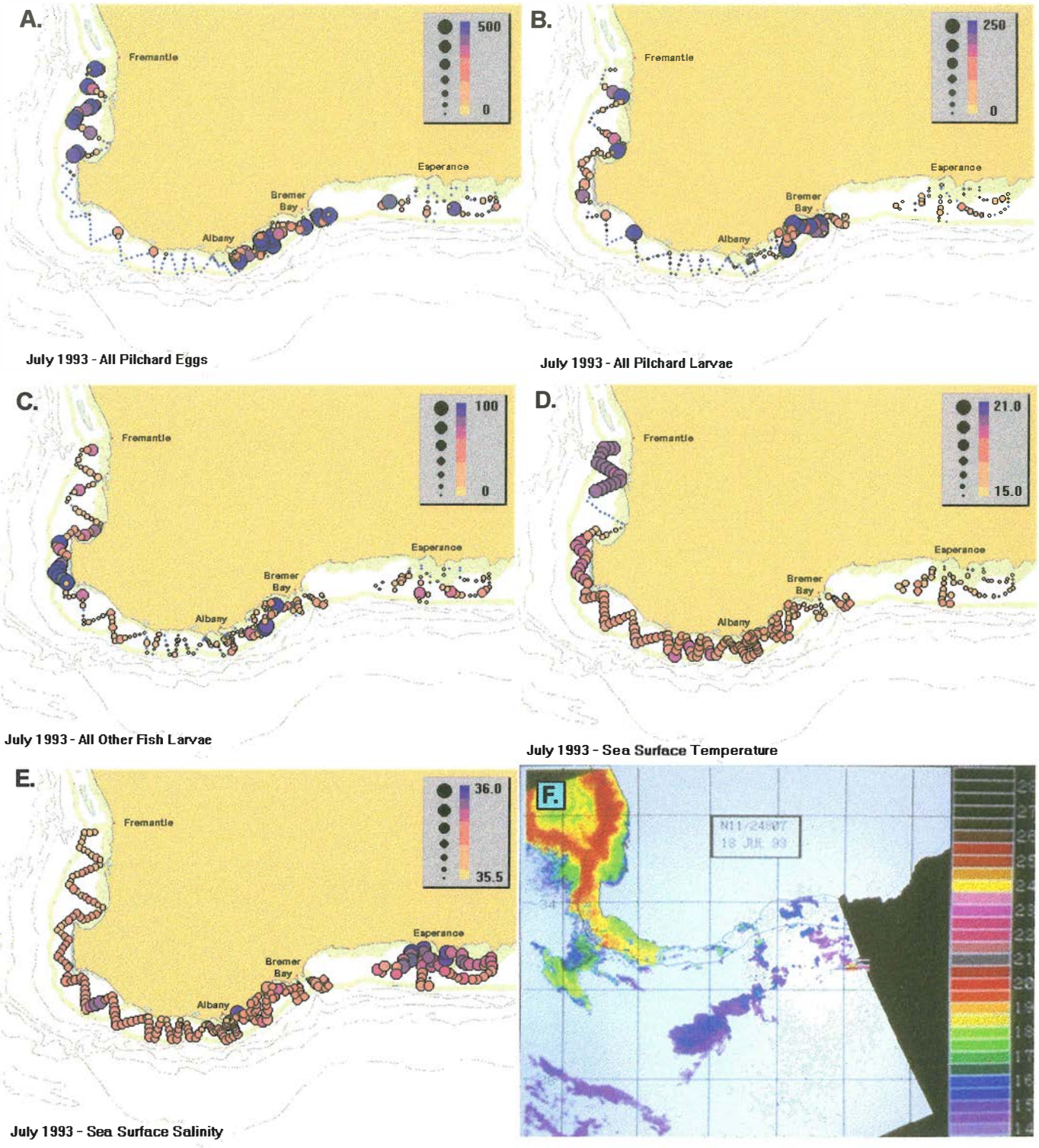


Figure 4.7: General plankton and physical data distributions during July 1993 field trip. **A.** *All pilchard eggs.* **B.** *All pilchard larvae.* **C.** *All other fish larvae.* **D.** *Sea surface temperature.* **E.** *Sea surface salinity.* **F.** *NOAA satellite image from 18/7/93 showing sea surface temperature.*

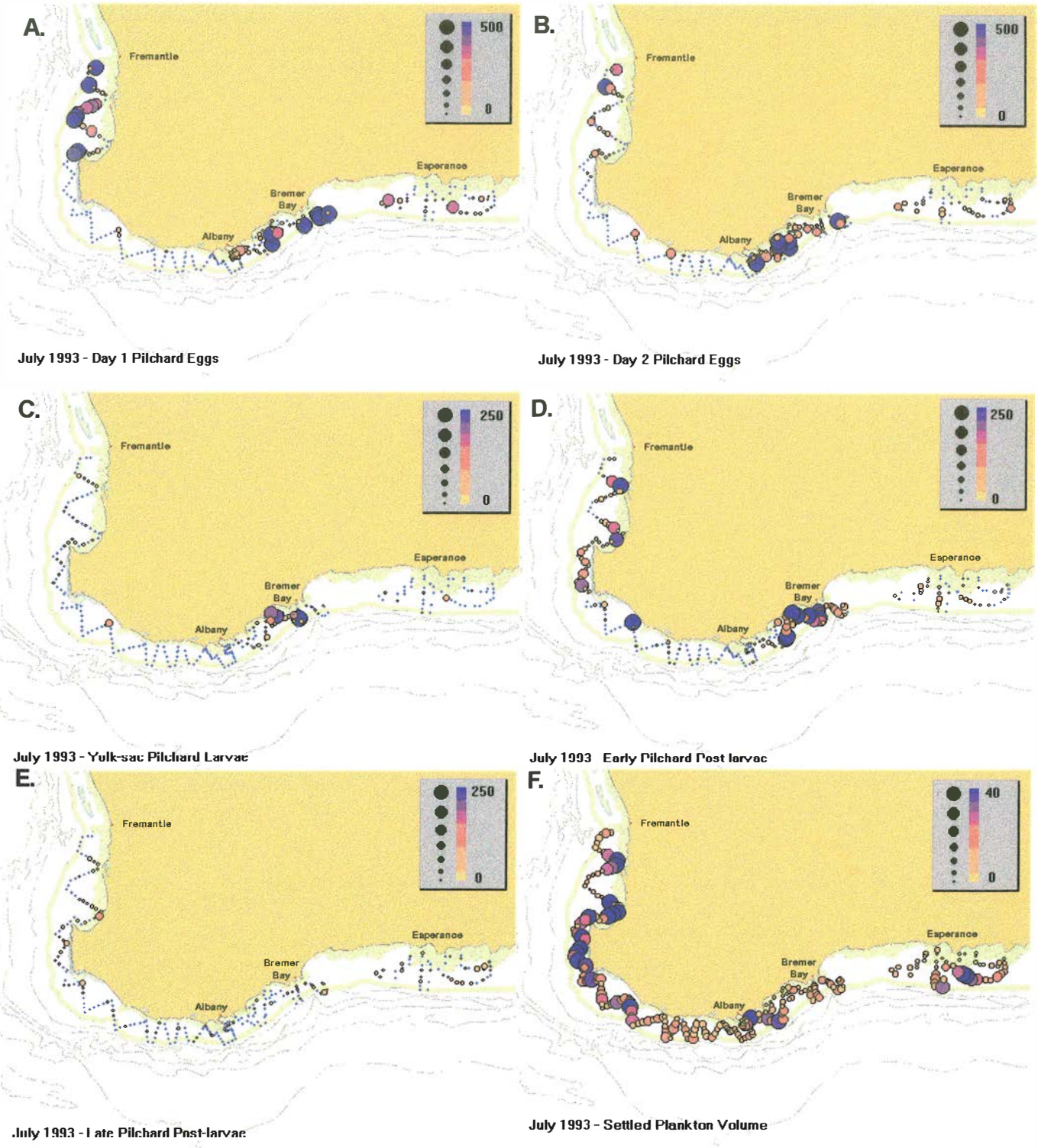


Figure 4.8: General plankton data distributions during July 1993 field trip.
A. *Day 1 pilchard eggs.* **B.** *Day 2 pilchard eggs.* **C.** *Yolk-sac pilchard larvae.*
D. *Early pilchard post-larvae.* **E.** *Late pilchard post-larvae.* **F.** *Settled plankton volume.*

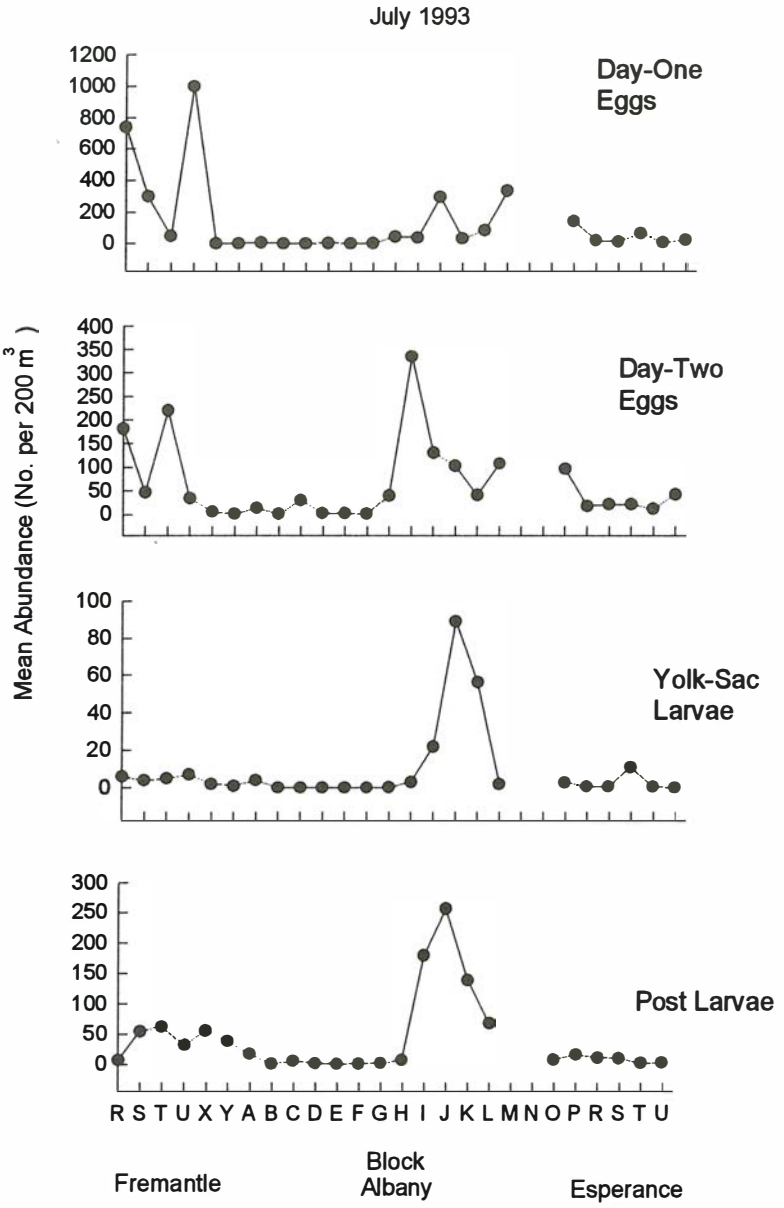


Figure 4.9: Relative distribution of pilchard stages Day-one, Day-two eggs, Yolk-sac and Post larvae collected in the July 1993 survey. The blocks refer to the sampling blocks detailed in Fig. 2.1.

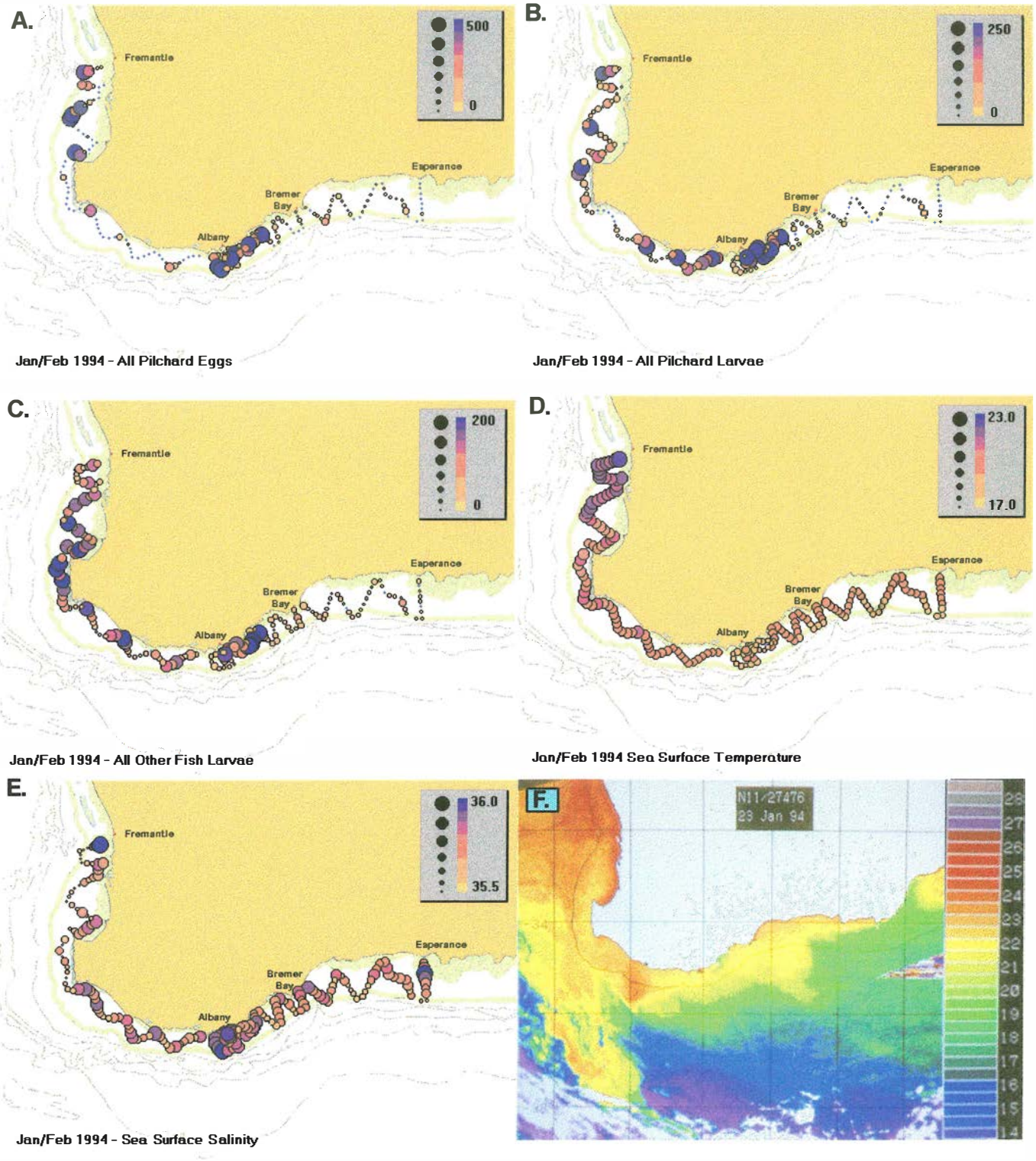


Figure 4.10: General plankton and physical data distributions during January 1994 field trip. **A.** *All pilchard eggs.* **B.** *All pilchard larvae.* **C.** *All other fish larvae.* **D.** *Sea surface temperature.* **E.** *Sea surface salinity.* **F.** *NOAA satellite image from 23-1-94 showing sea surface temperature.*

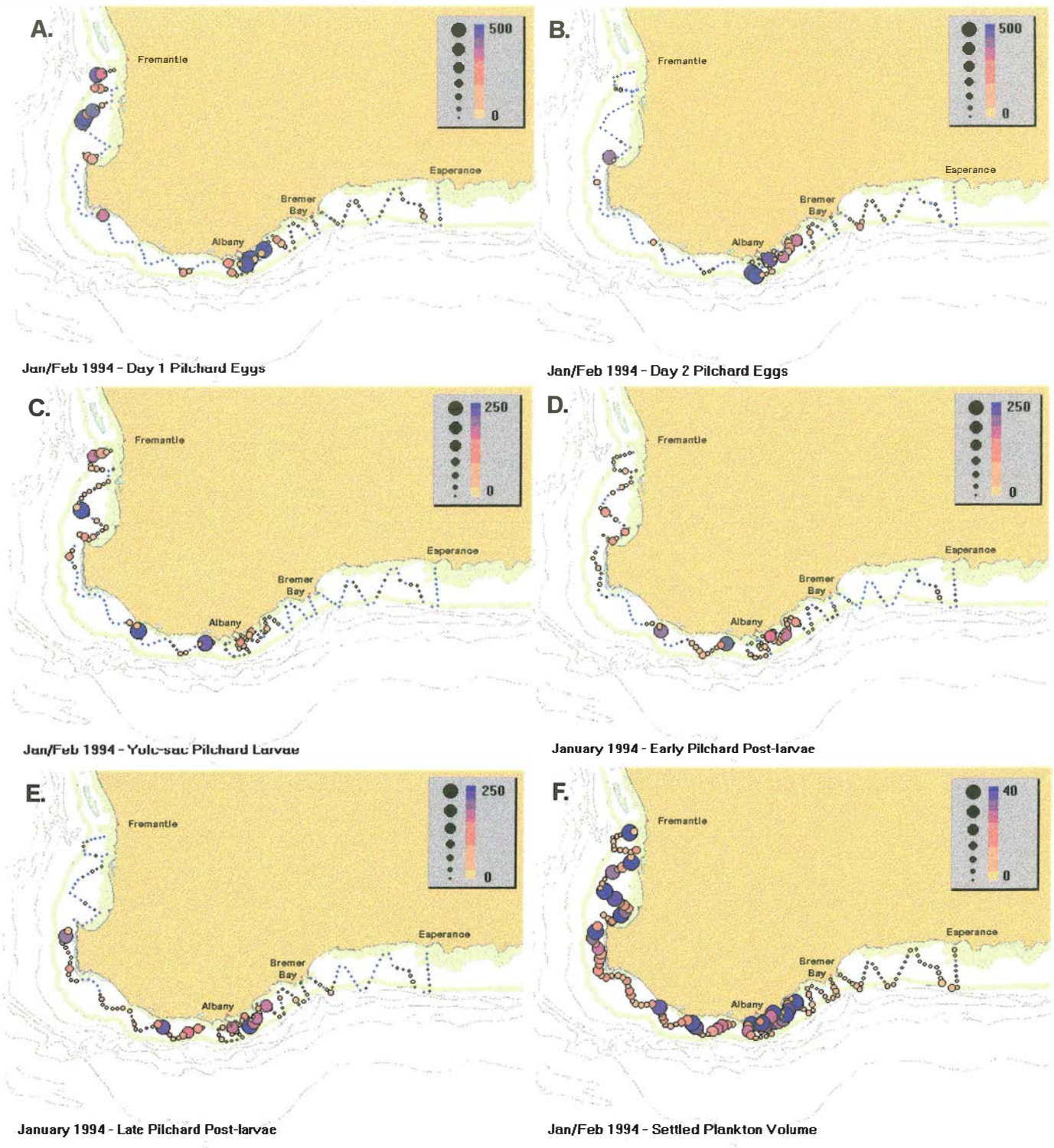


Figure 4.11: General plankton data distributions during January 1994 field trip.
A. *Day 1 pilchard eggs.* **B.** *Day 2 pilchard eggs.* **C.** *Yolk-sac pilchard larvae.*
D. *Early pilchard post-larvae.* **E.** *Late pilchard post-larvae.* **F.** *Settled plankton volume.*

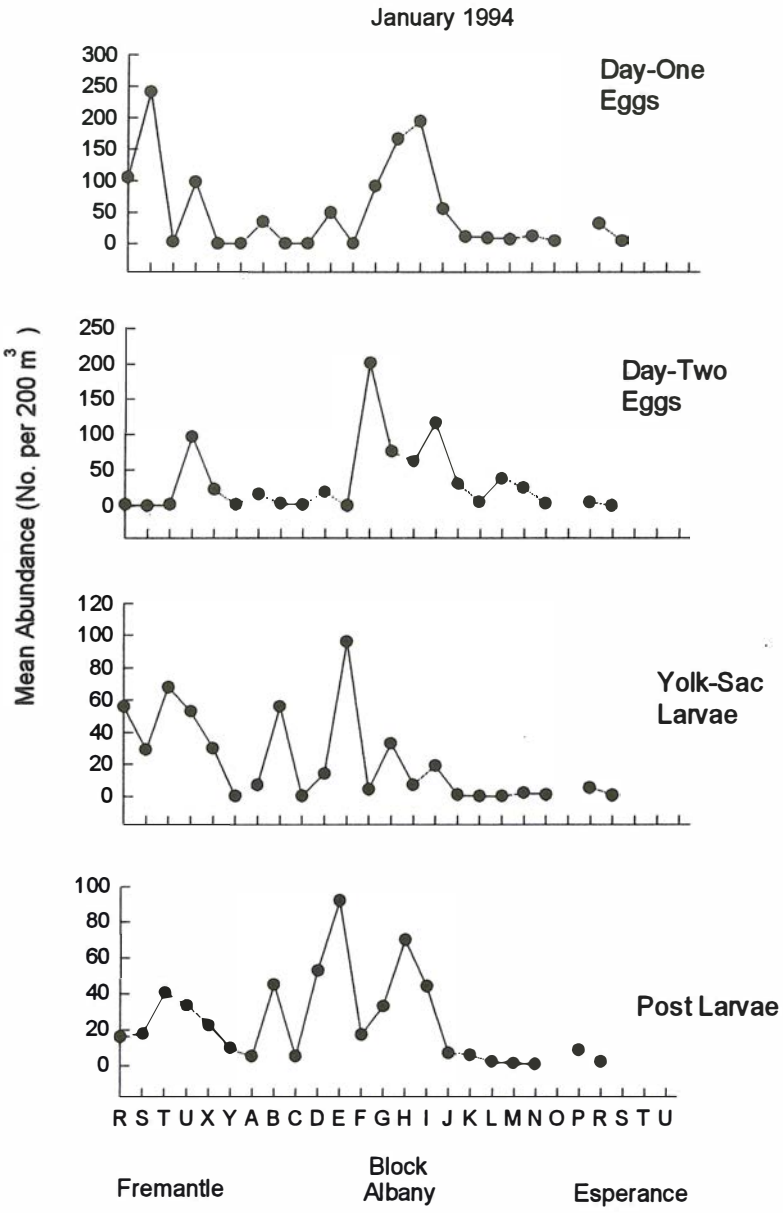
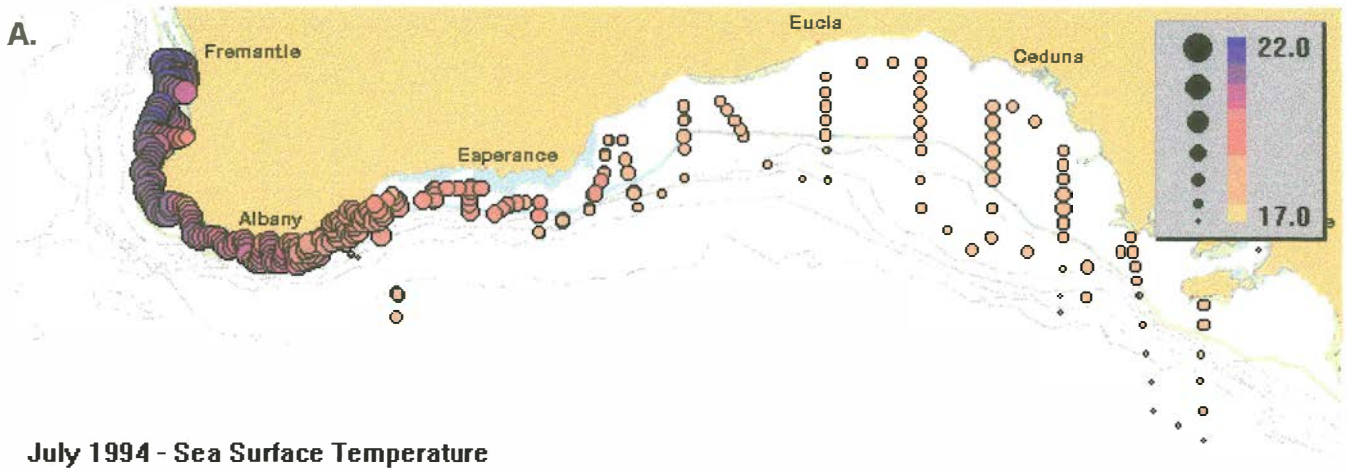
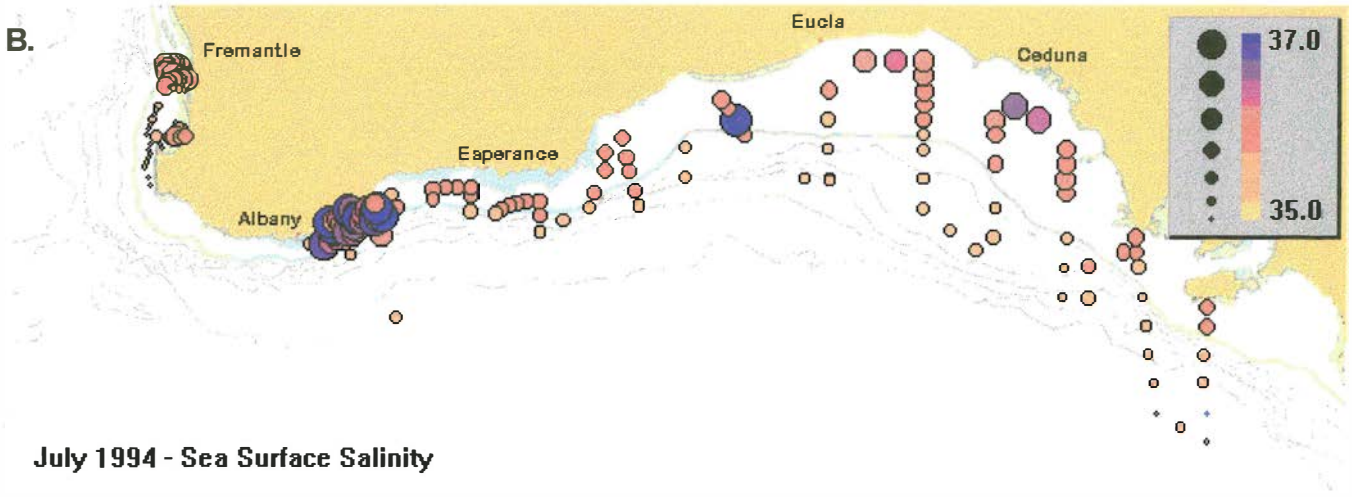


Figure 4.12: Relative distribution of pilchard stages Day-one, Day-two eggs, Yolk-sac and post larvae collected in the January 1994 survey. The blocks refer to the sampling blocks detailed in Fig. 2.1.





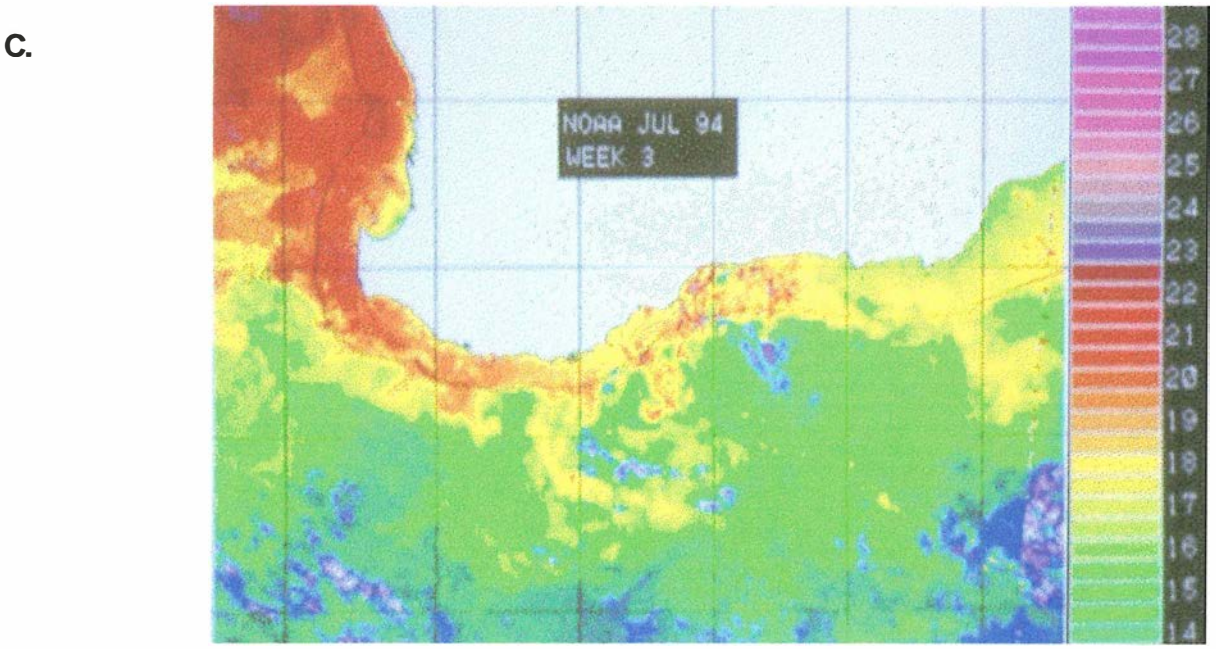


Figure 4.13: General physical data - July 1994 field trip.
A. *Sea Surface Temperature.* **B.** *Sea Surface Salinity*. **C.** *NOAA satellite photo taken week 3 July 1994*

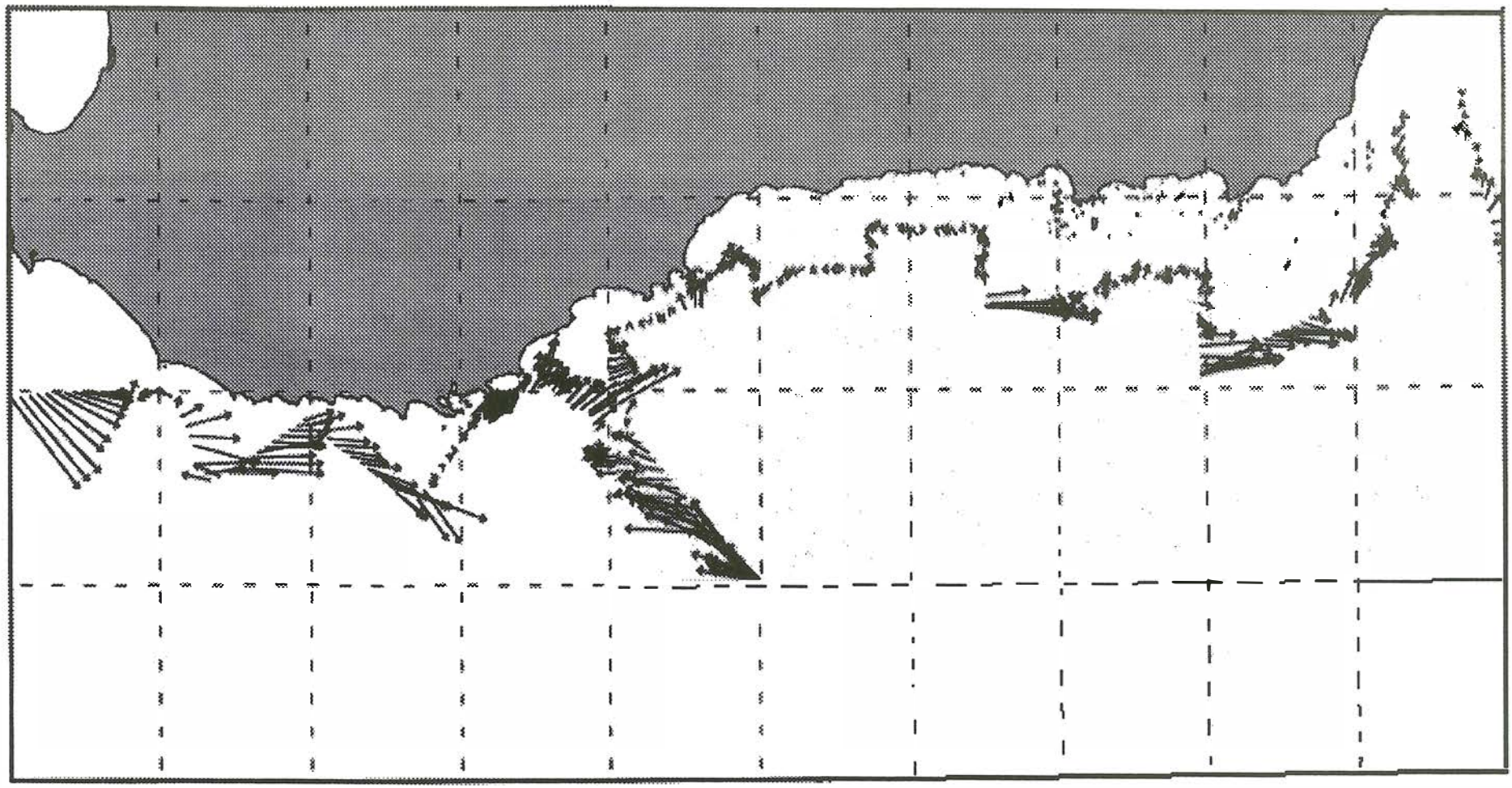
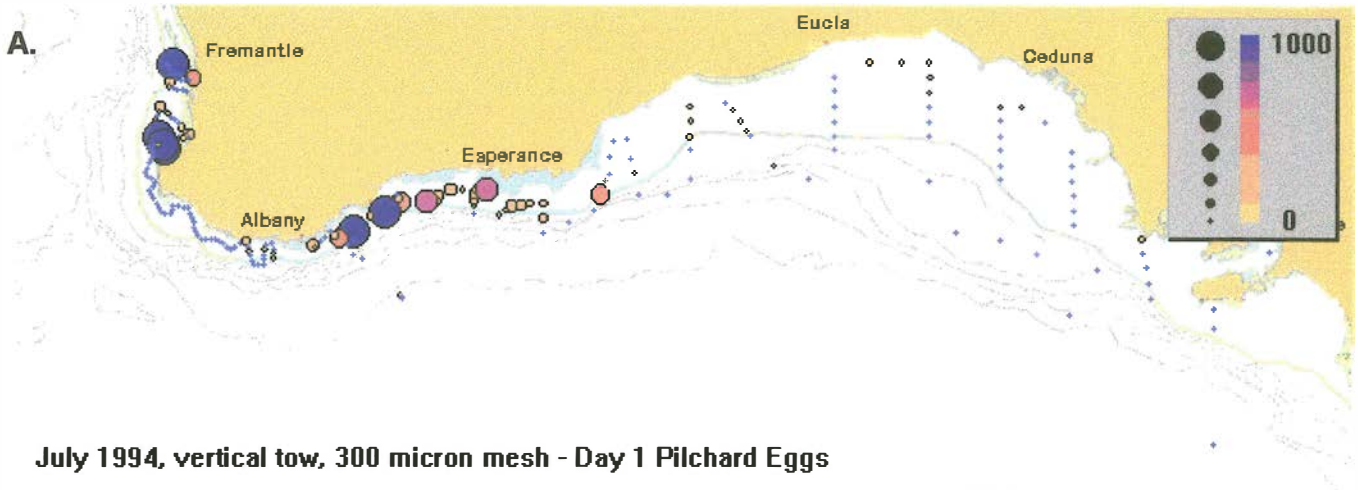
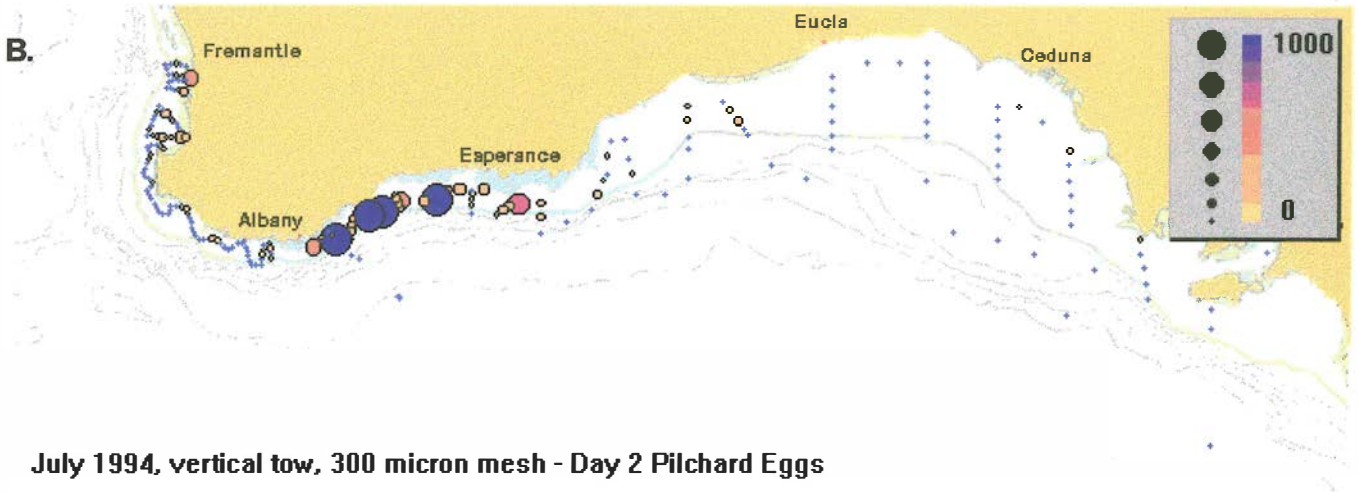


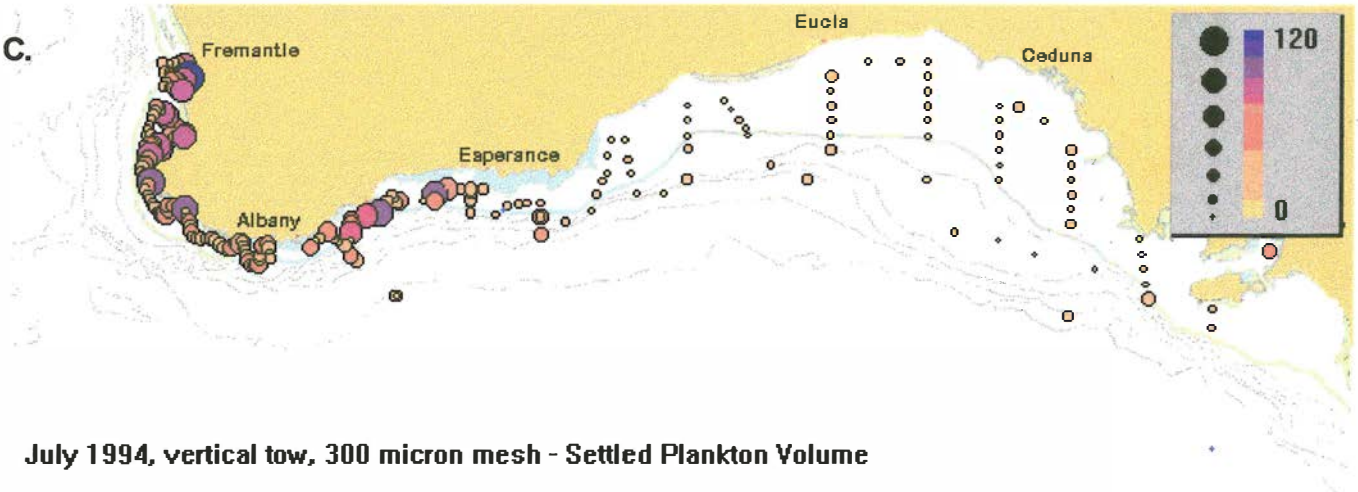
Figure 4.14: ADCP measurements taken aboard the *R.V. Franklin* during July 1994 plankton survey.



July 1994, vertical tow, 300 micron mesh - Day 1 Pilchard Eggs



July 1994, vertical tow, 300 micron mesh - Day 2 Pilchard Eggs



July 1994, vertical tow, 300 micron mesh - Settled Plankton Volume

Figure 4.15: General plankton distribution with 300 micron mesh, vertical tow - July 1994 field trip. **A.** *Day 1 pilchard eggs.* **B.** *Day 2 pilchard eggs.* **C.** *Settled plankton volume . .*

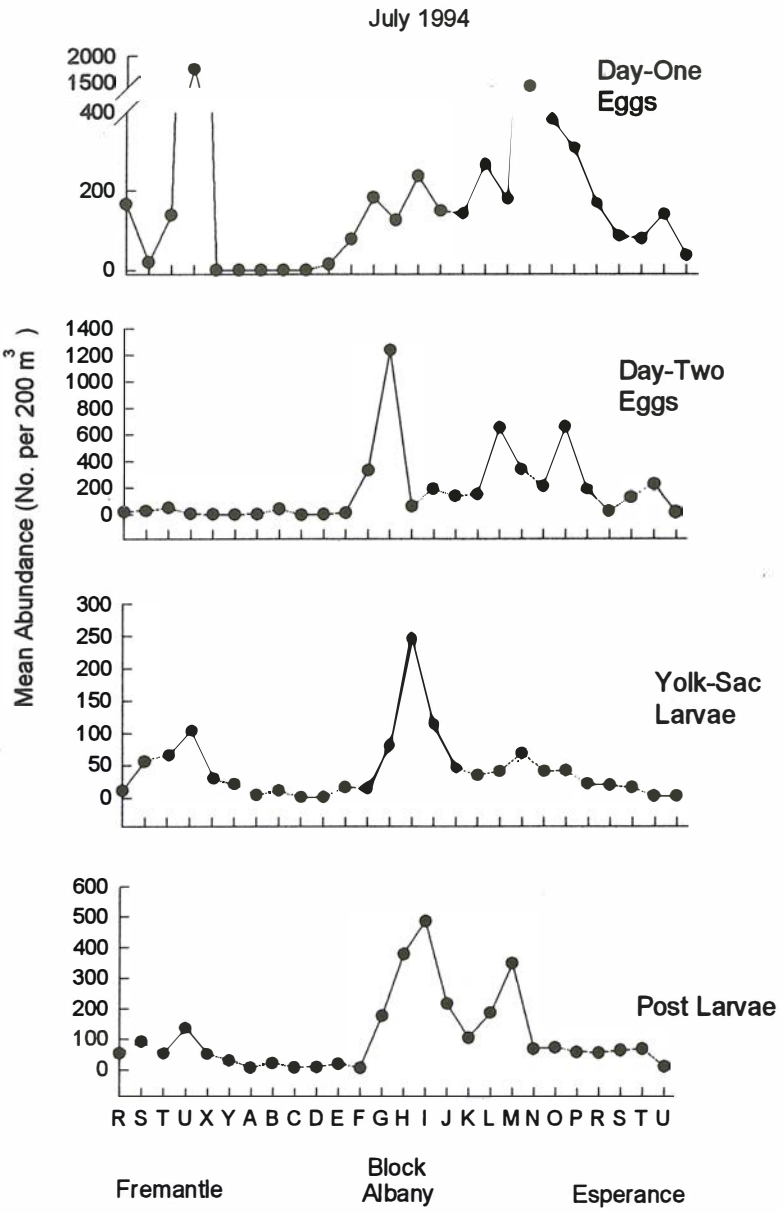
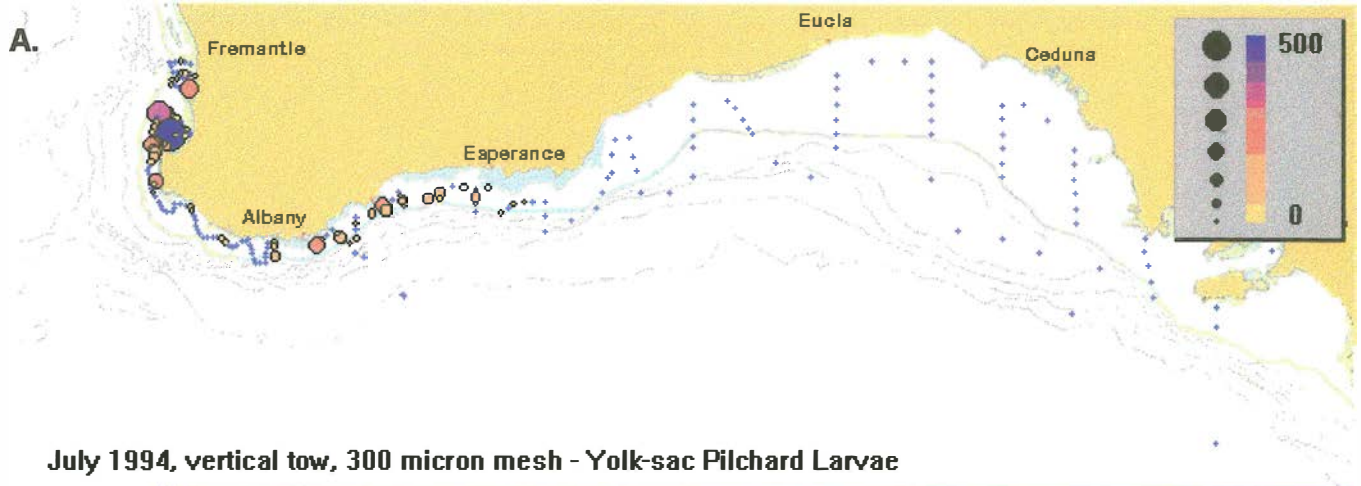
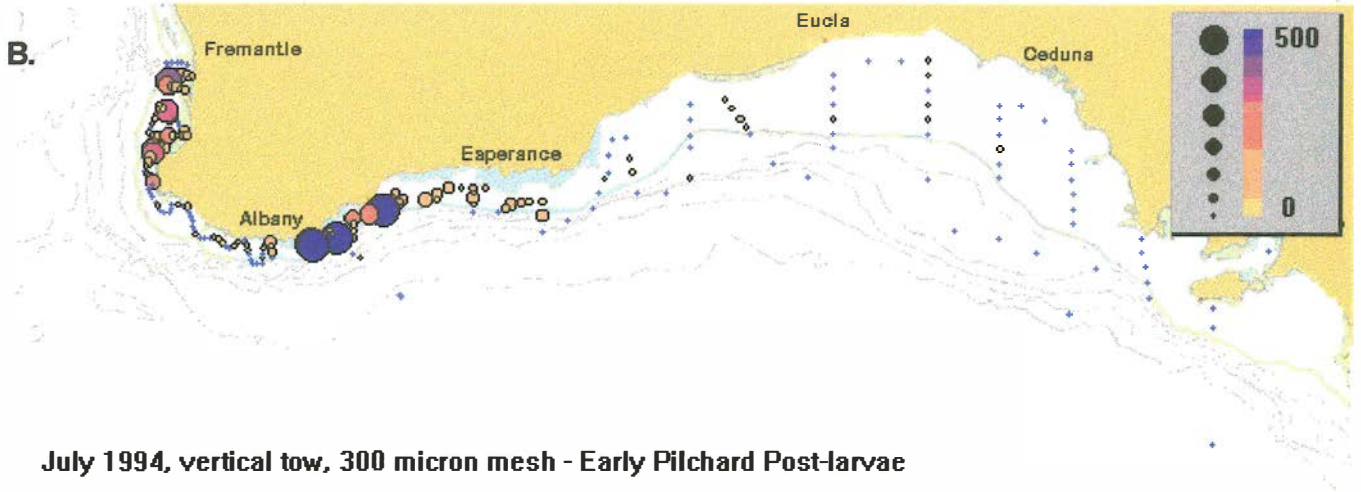


Figure 4.16: Relative distribution of pilchard stages Day-one, Day-two eggs, Yolk-sac and post larvae collected in the July 1994 survey. The blocks refer to the sampling blocks detailed in Fig. 2.1.





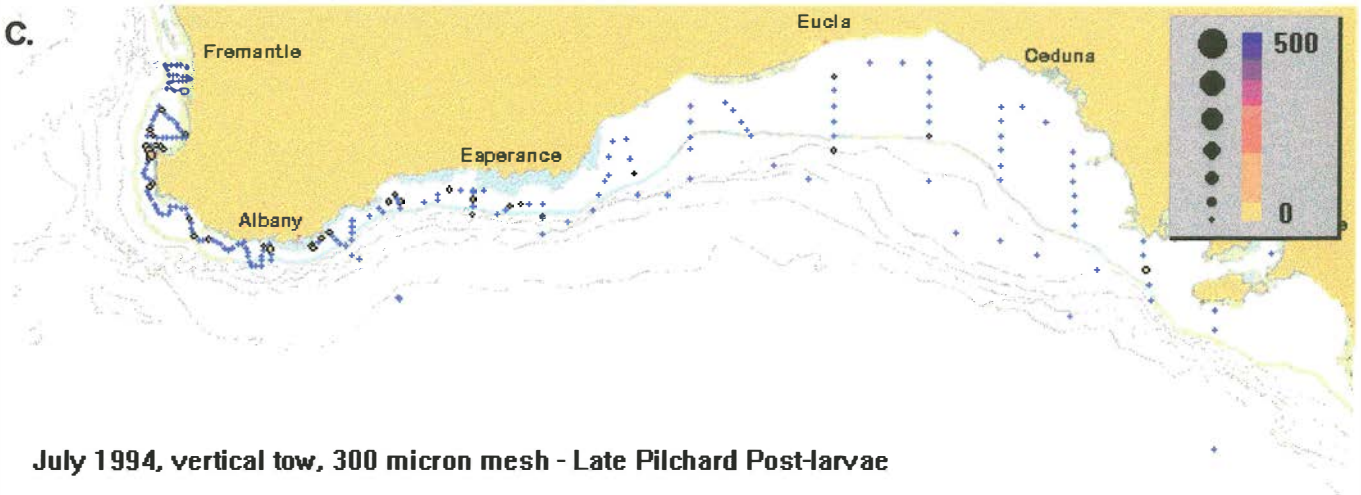
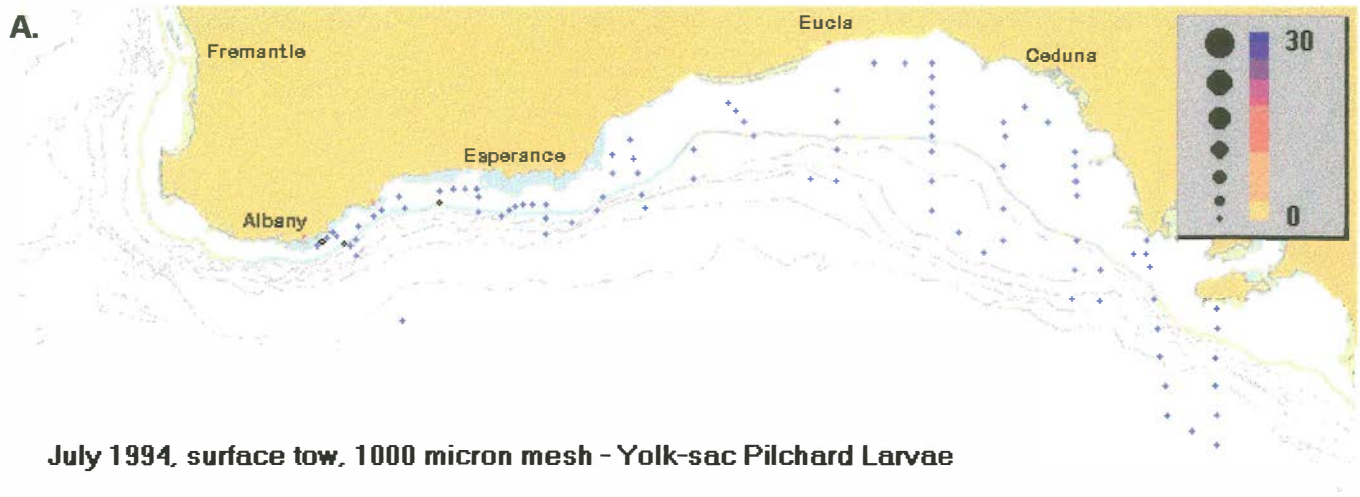
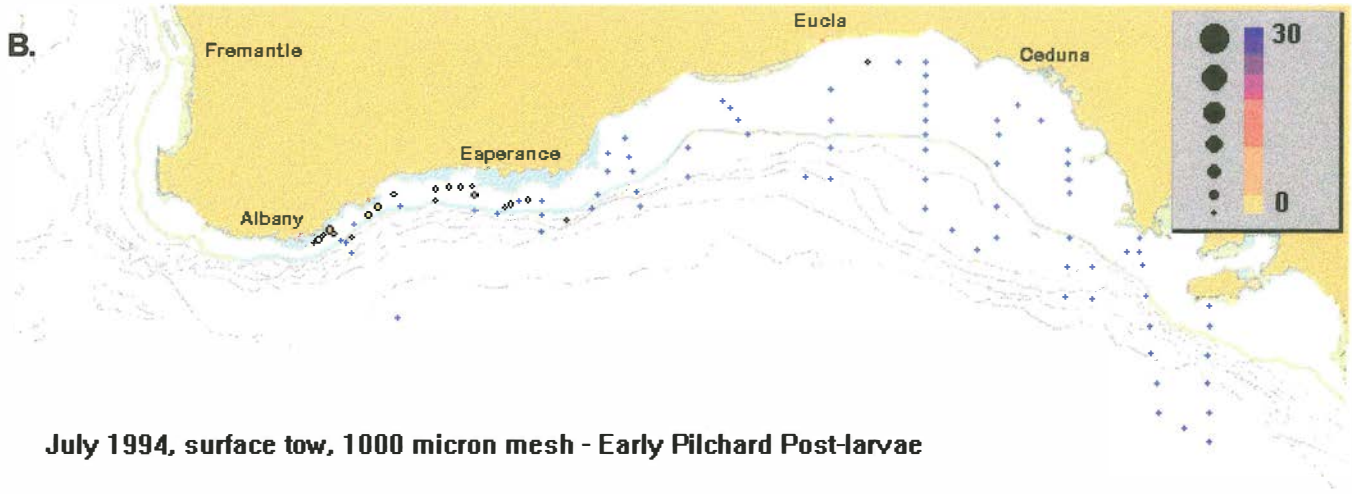


Figure 4.17: General plankton distribution with 300 micron mesh, vertical tow - July 1994 field trip. **A.** *Yolk-sac pilchard larvae.* **B.** *Early pilchard post-larvae.* **C.** *Late pilchard post-larvae.* .





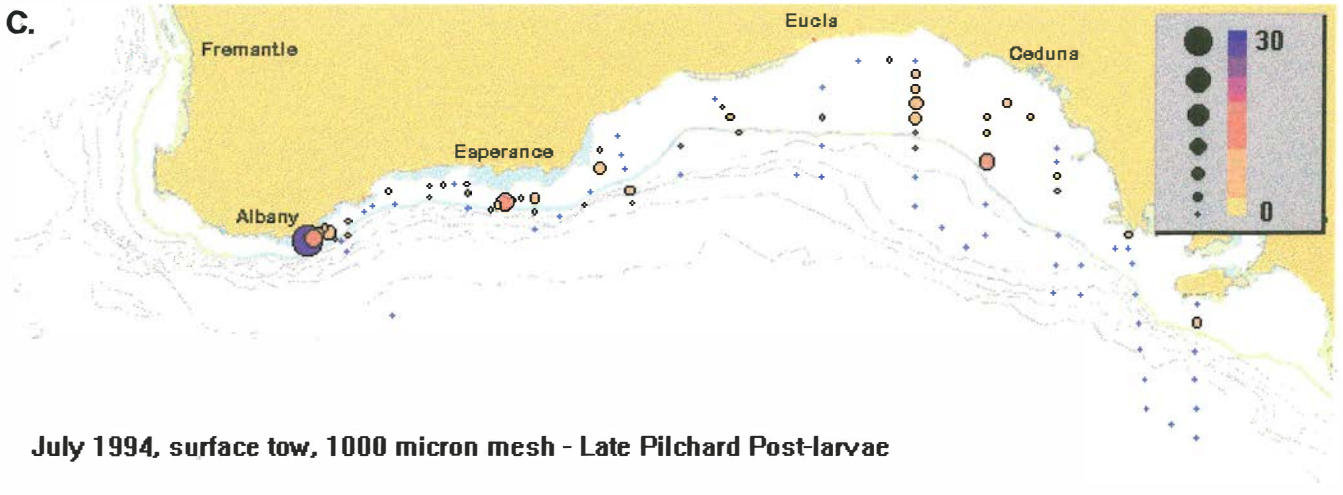
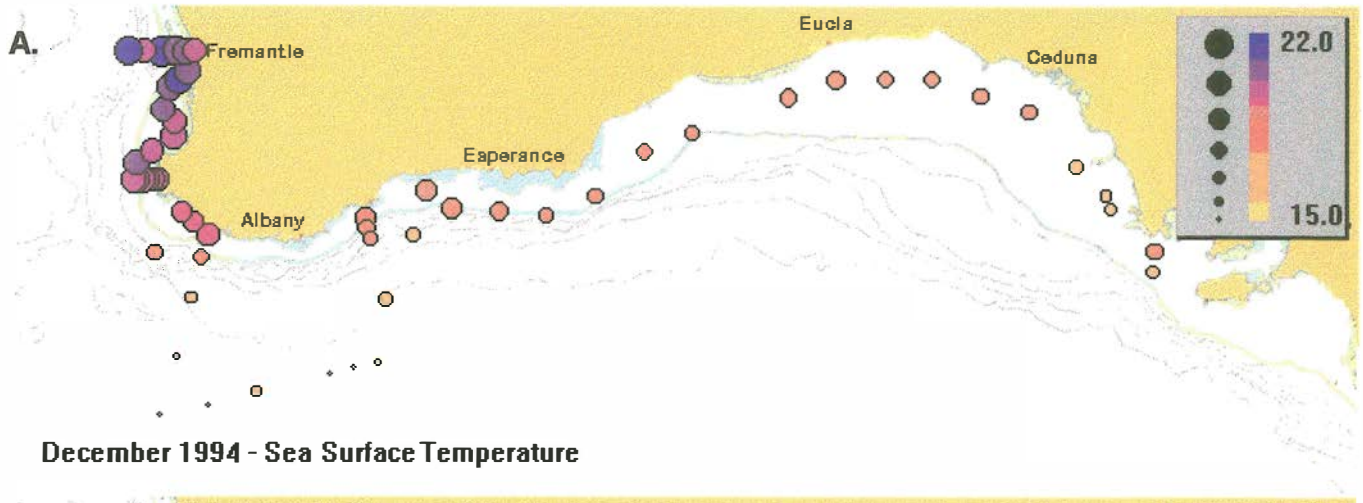
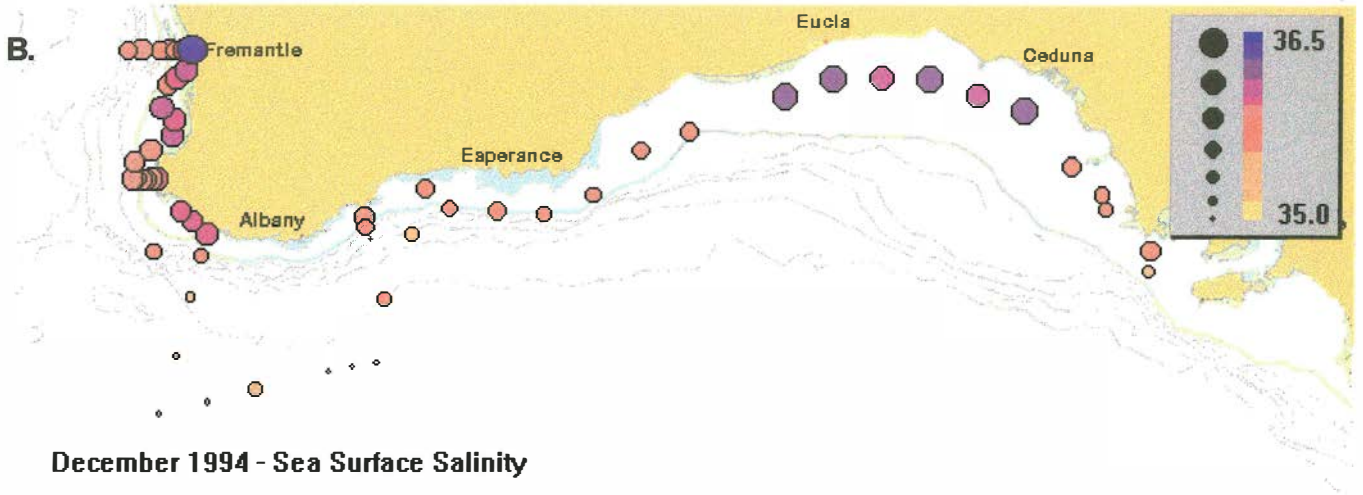


Figure 4.18: General plankton distribution using 1000 micron mesh surface net - July 1994 field trip. **A.** *Yolk-sac pilchard larvae.* **B.** *Early pilchard post-larvae.* **C.** *Late pilchard post-larvae..*





C.

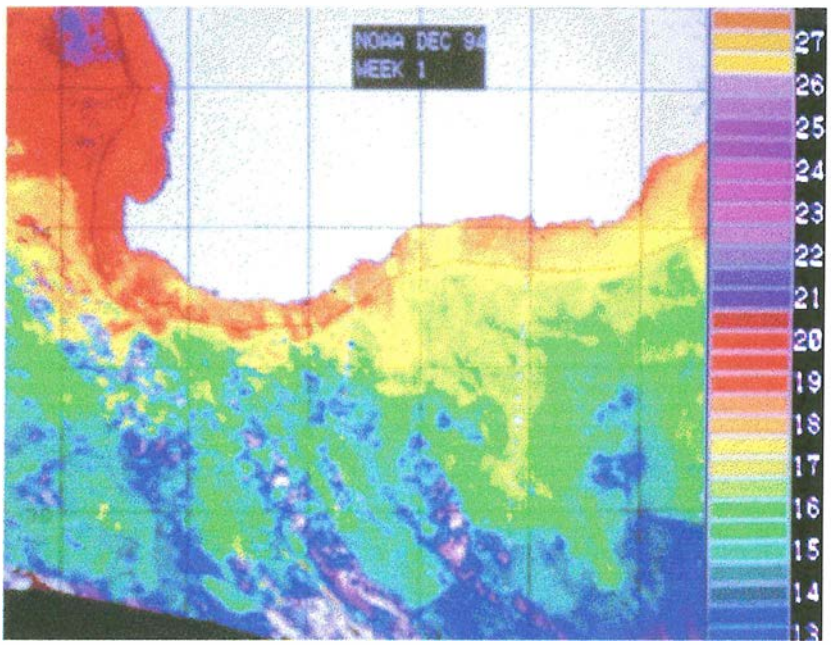
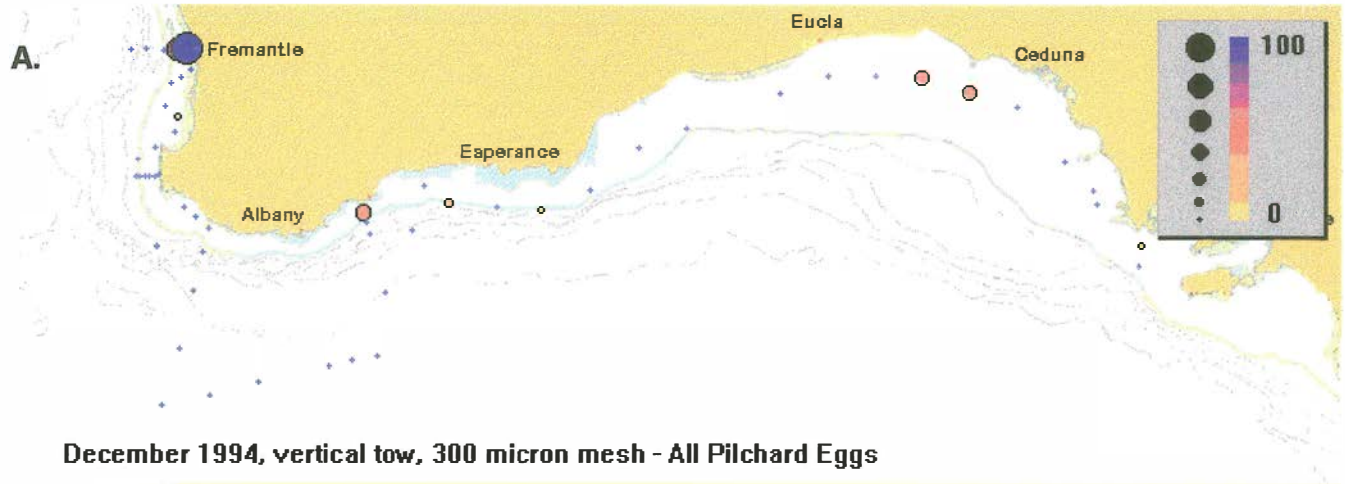
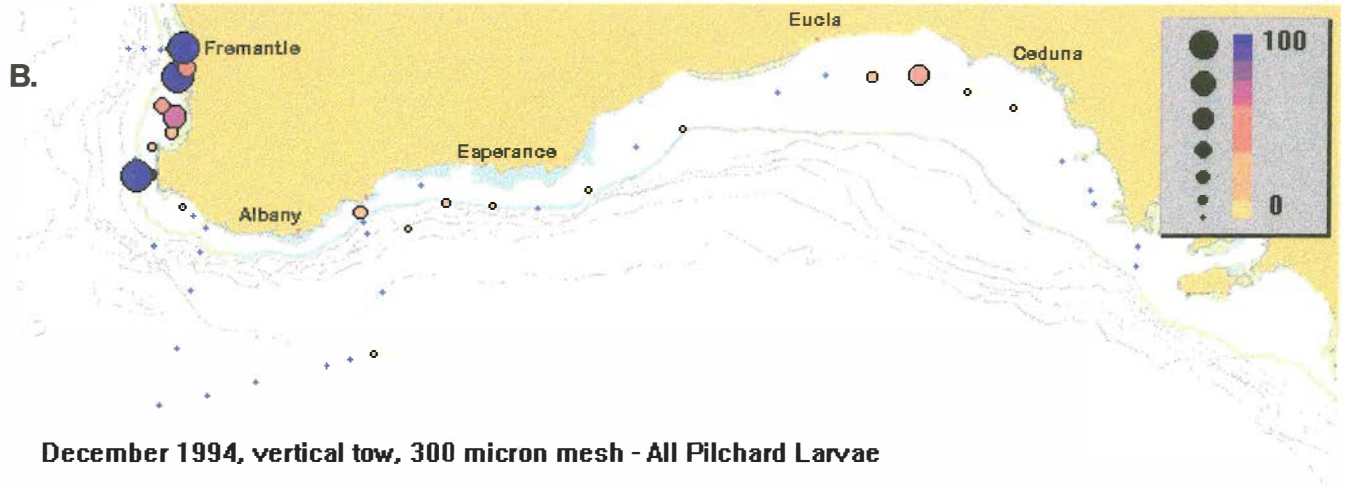


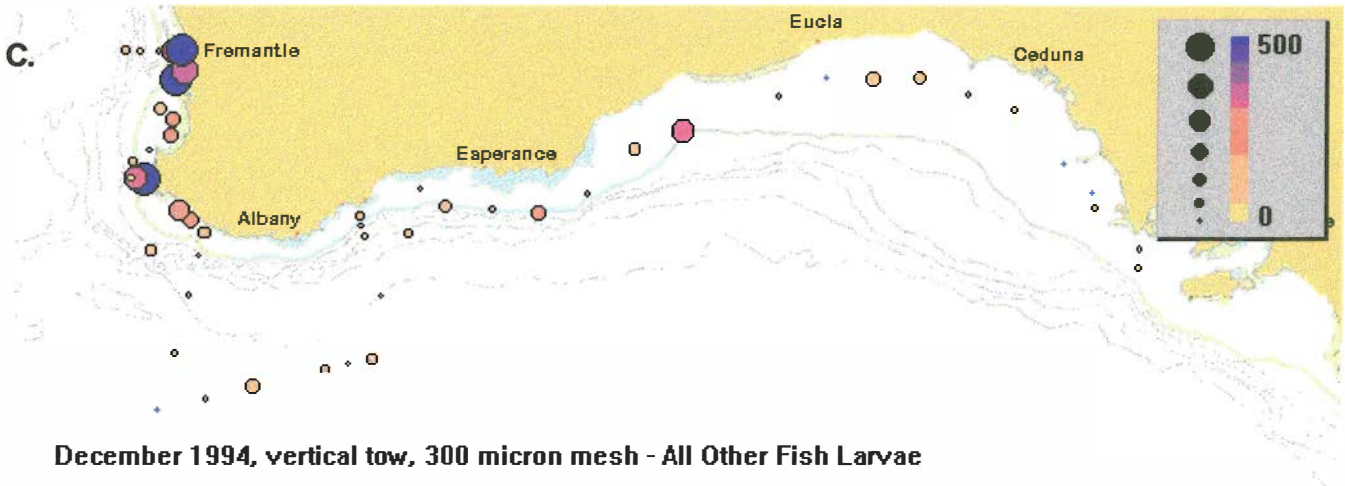
Figure 4.19: General physical data - December 1994 field trip.
A. *Sea Surface Temperature.* **B.** *Sea Surface Salinity*. **C.** *NOAA satellite photo taken week 3 July 1994*



December 1994, vertical tow, 300 micron mesh - All Pilchard Eggs

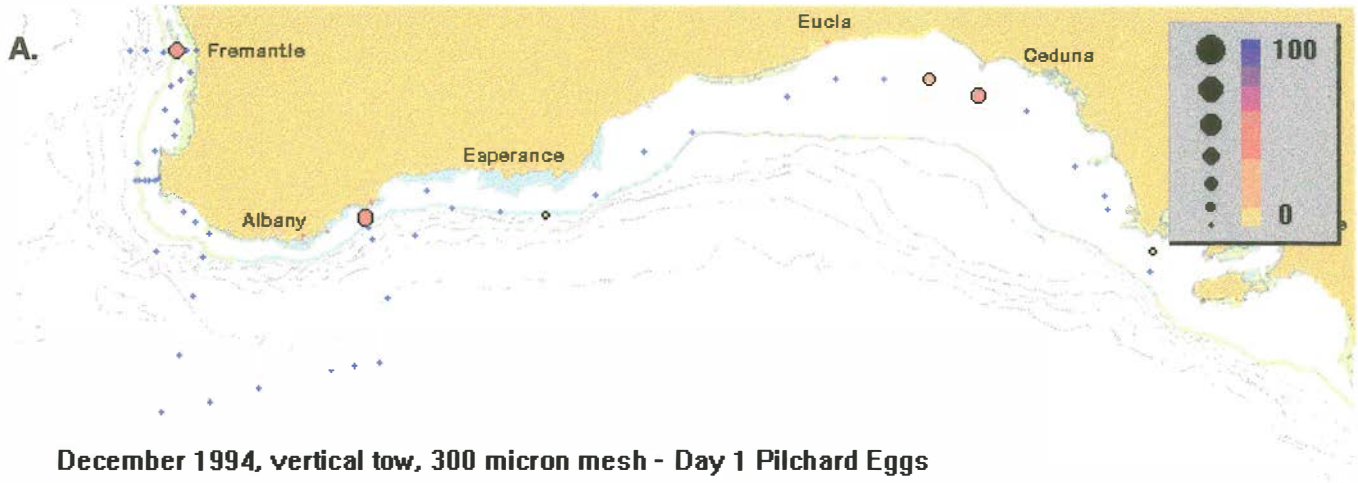


December 1994, vertical tow, 300 micron mesh - All Pilchard Larvae

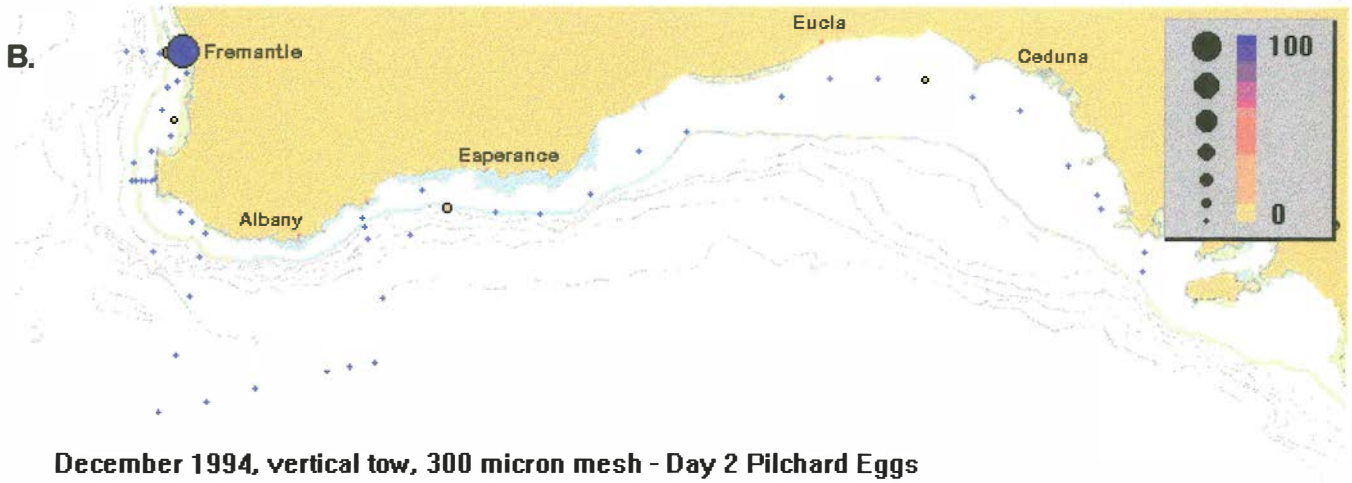


December 1994, vertical tow, 300 micron mesh - All Other Fish Larvae

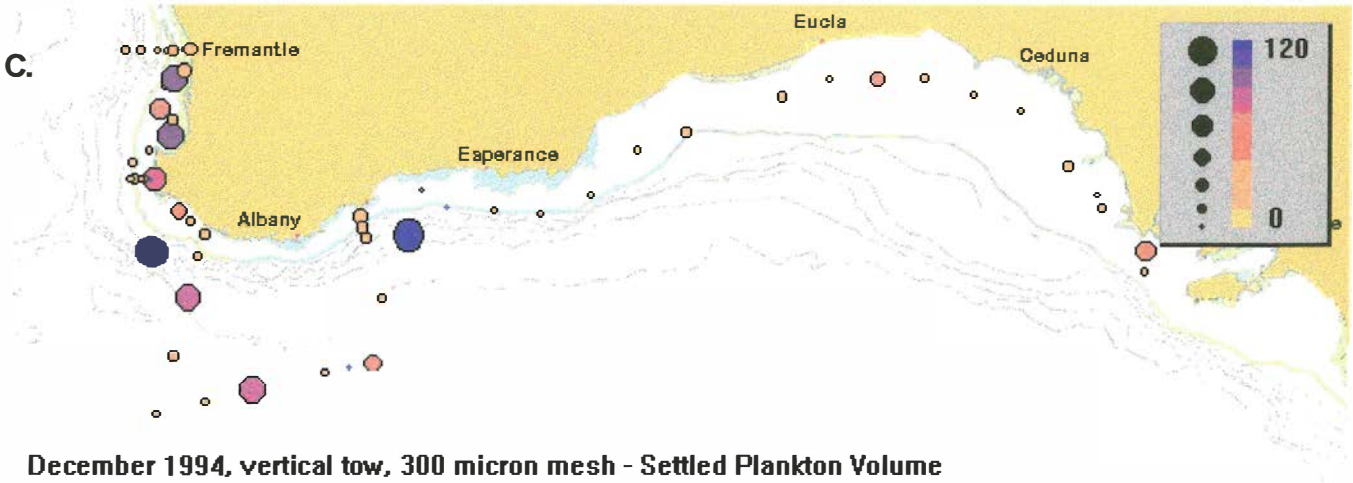
Figure 4.20: General plankton distribution with 300 micron mesh, vertical tows - December 1994 field trip. **A.** *All pilchard eggs.* **B.** *All pilchard larvae.* **C.** *All other fish larvae.*



December 1994, vertical tow, 300 micron mesh - Day 1 Pilchard Eggs

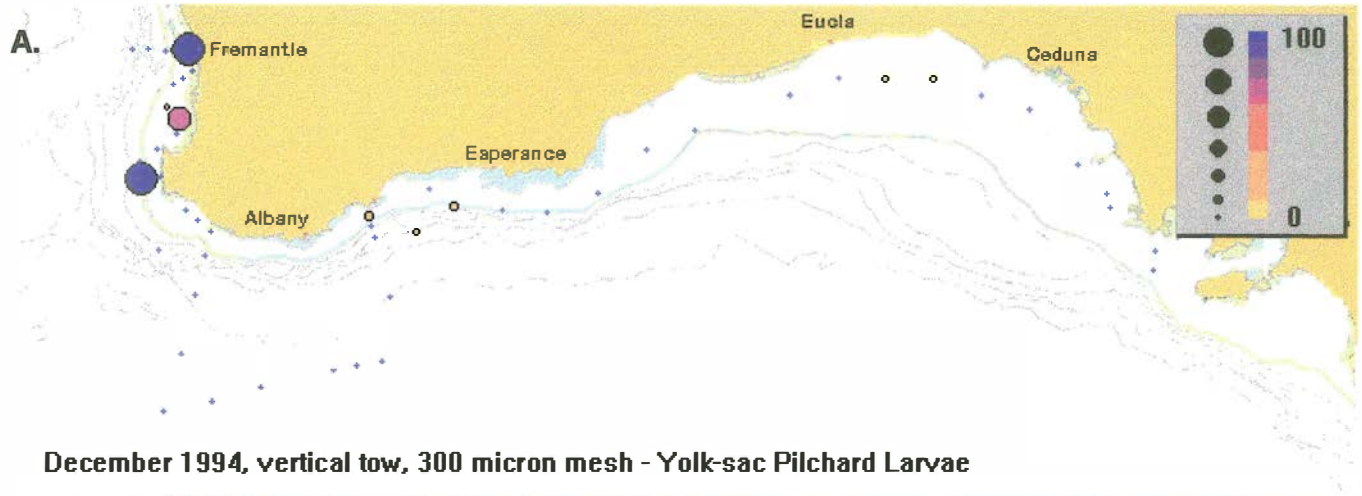


December 1994, vertical tow, 300 micron mesh - Day 2 Pilchard Eggs

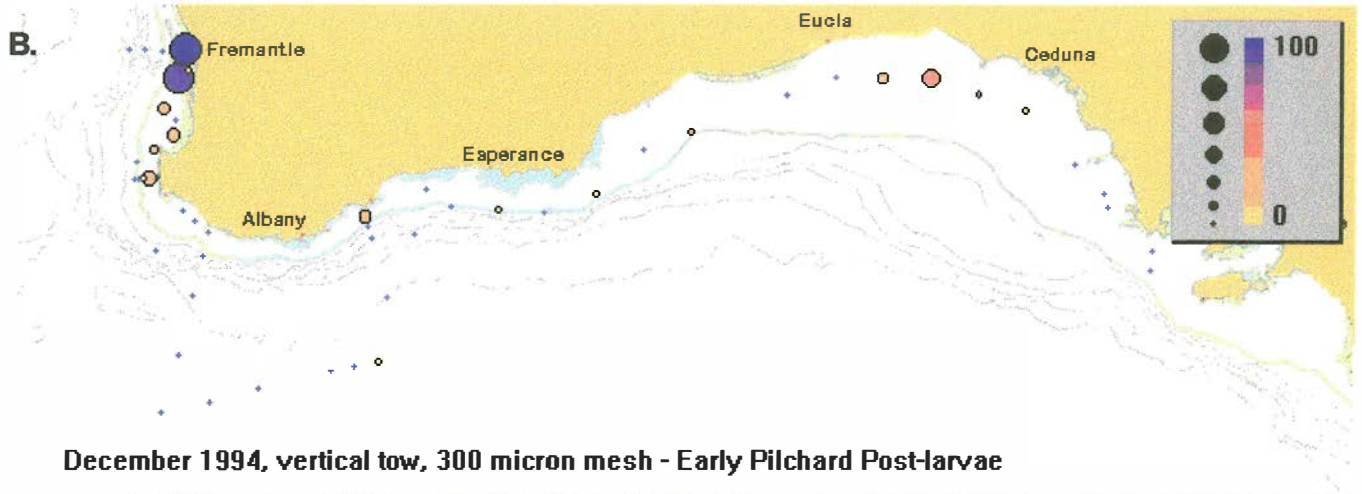


December 1994, vertical tow, 300 micron mesh - Settled Plankton Volume

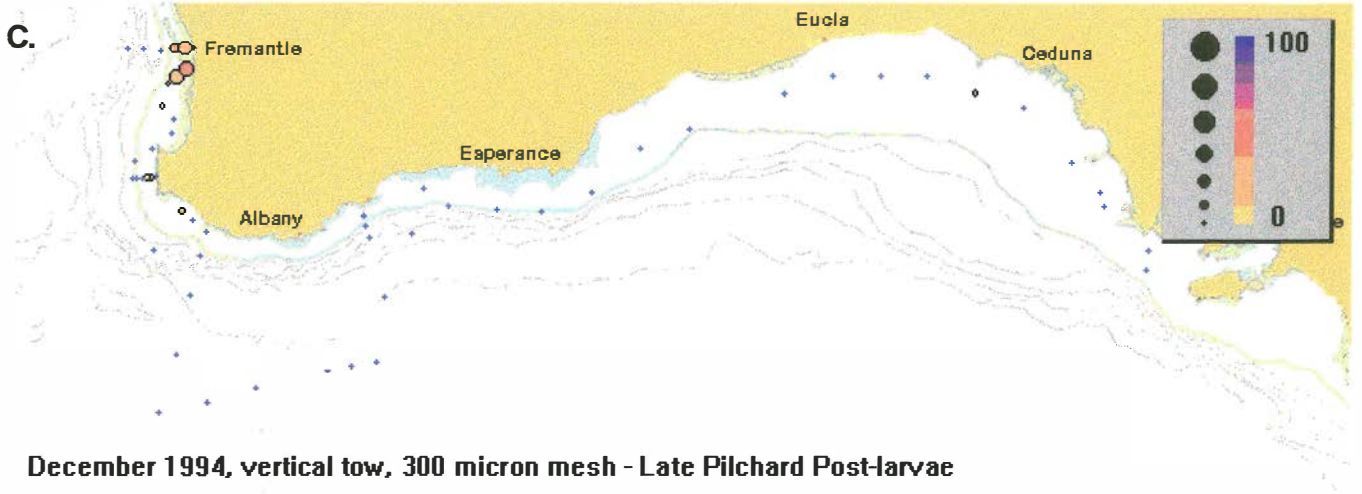
Figure 4.21: General plankton distribution with 300 micron mesh, vertical tow - December 1994 field trip. **A.** *Day 1 pilchard eggs.* **B.** *Day 2 pilchard eggs.* **C.** *Settled plankton volume . .*



December 1994, vertical tow, 300 micron mesh - Yolk-sac Pilchard Larvae

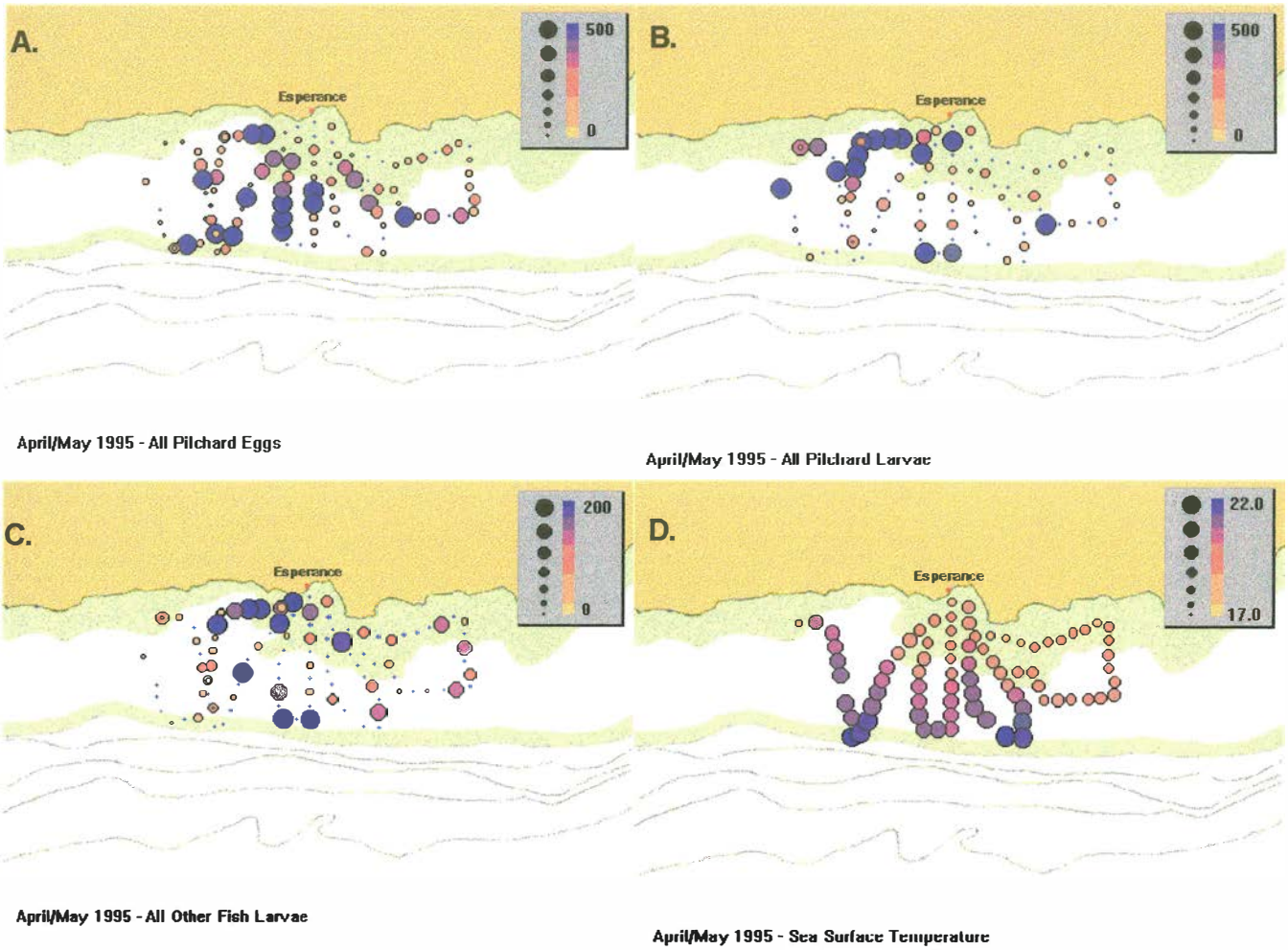


December 1994, vertical tow, 300 micron mesh - Early Pilchard Post-larvae



December 1994, vertical tow, 300 micron mesh - Late Pilchard Post-larvae

Figure 4.22: General plankton distribution with 300 micron mesh, vertical tow - December 1994 field trip. **A.** *Yolk-sac pilchard larvae.* **B.** *Early pilchard post-larvae.* **C.** *Late pilchard post-larvae.* .



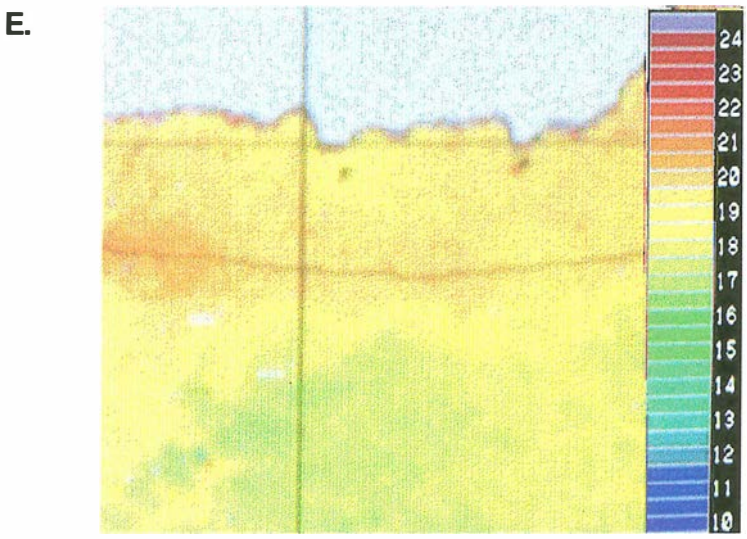


Figure 4.23: General plankton and physical data distributions during April/May 1995 field trip. **A.** *All pilchard eggs.* **B.** *All pilchard larvae.* **C.** *All other fish larvae.* **D.** *Sea surface temperature.* **E.** *NOAA satellite image from week 1 May 1995 showing sea surface temperature.*

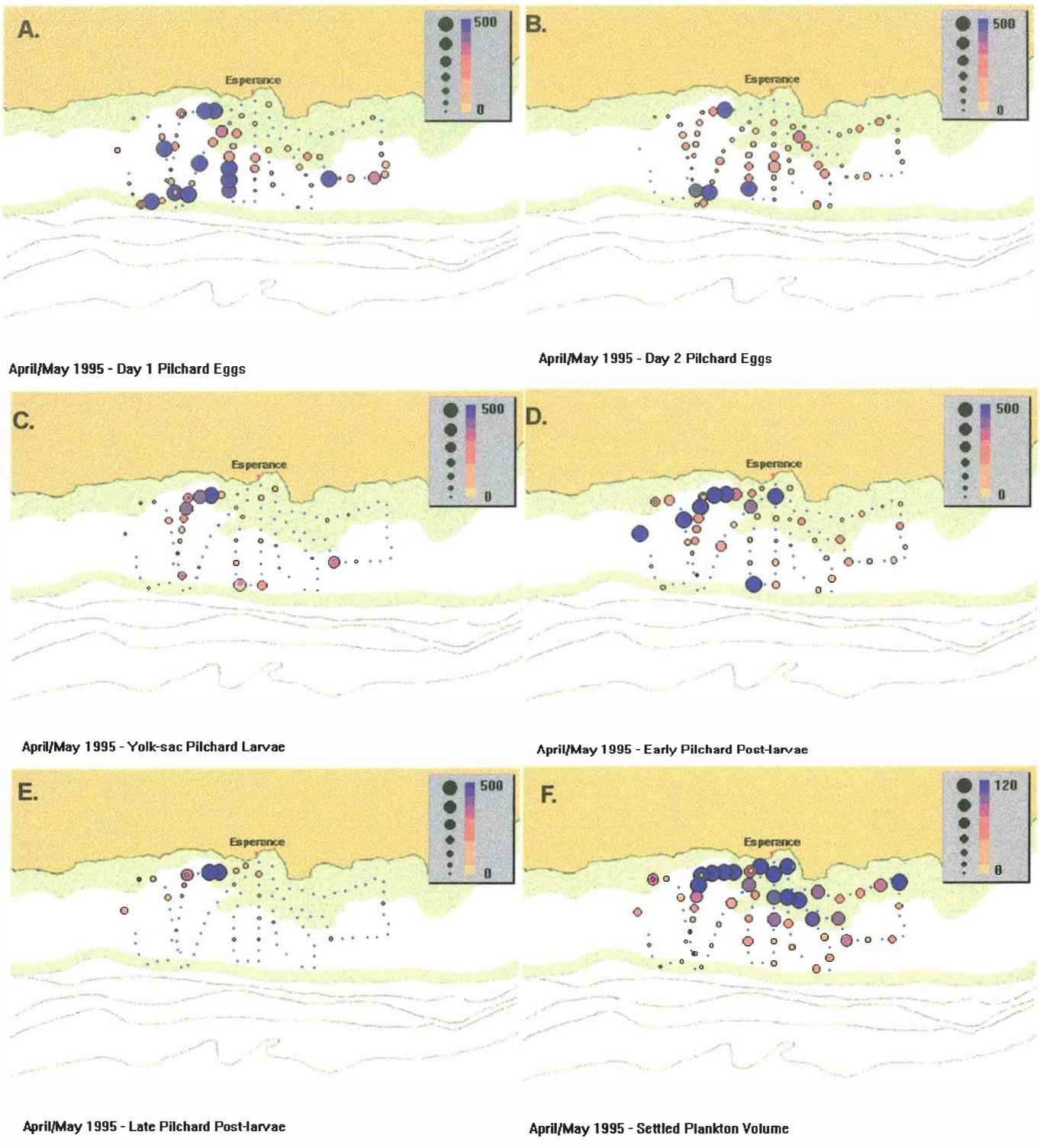


Figure 4.24: General plankton data distributions during April/May 1995 field trip.
A. *Day 1 pilchard eggs.* **B.** *Day 2 pilchard eggs.* **C.** *Yolk-sac pilchard larvae.*
D. *Early pilchard post-larvae.* **E.** *Late pilchard post-larvae.* **F.** *Settled plankton volume.*

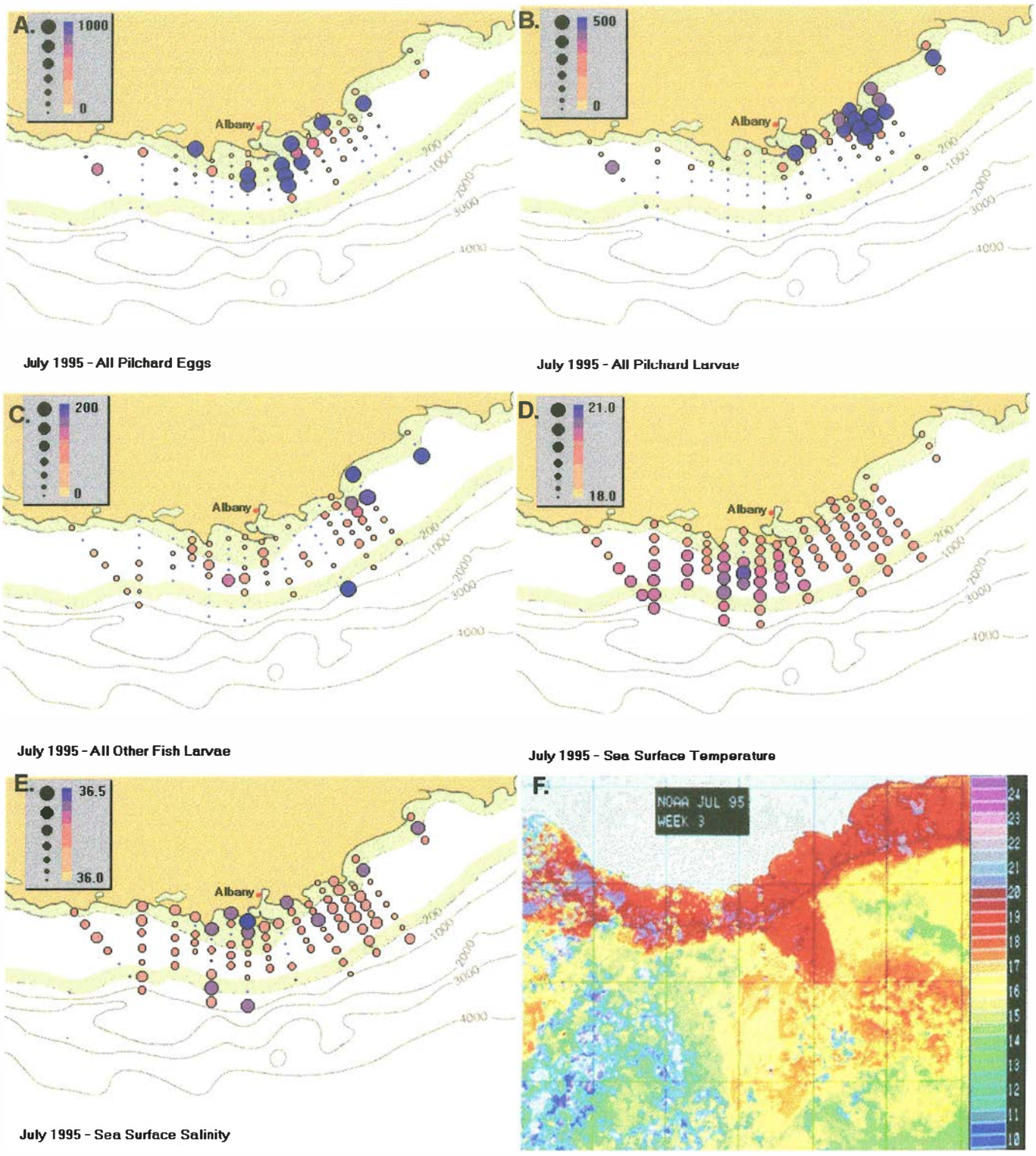


Figure 4.25: General plankton and physical data distributions during July 1995 field trip. **A.** *All pilchard eggs.* **B.** *All pilchard larvae.* **C.** *All other fish larvae.* **D.** *Sea surface temperature.* **E.** *Sea surface salinity.* **F.** *NOAA satellite image from July 1995, week 3 showing sea surface temperature.*

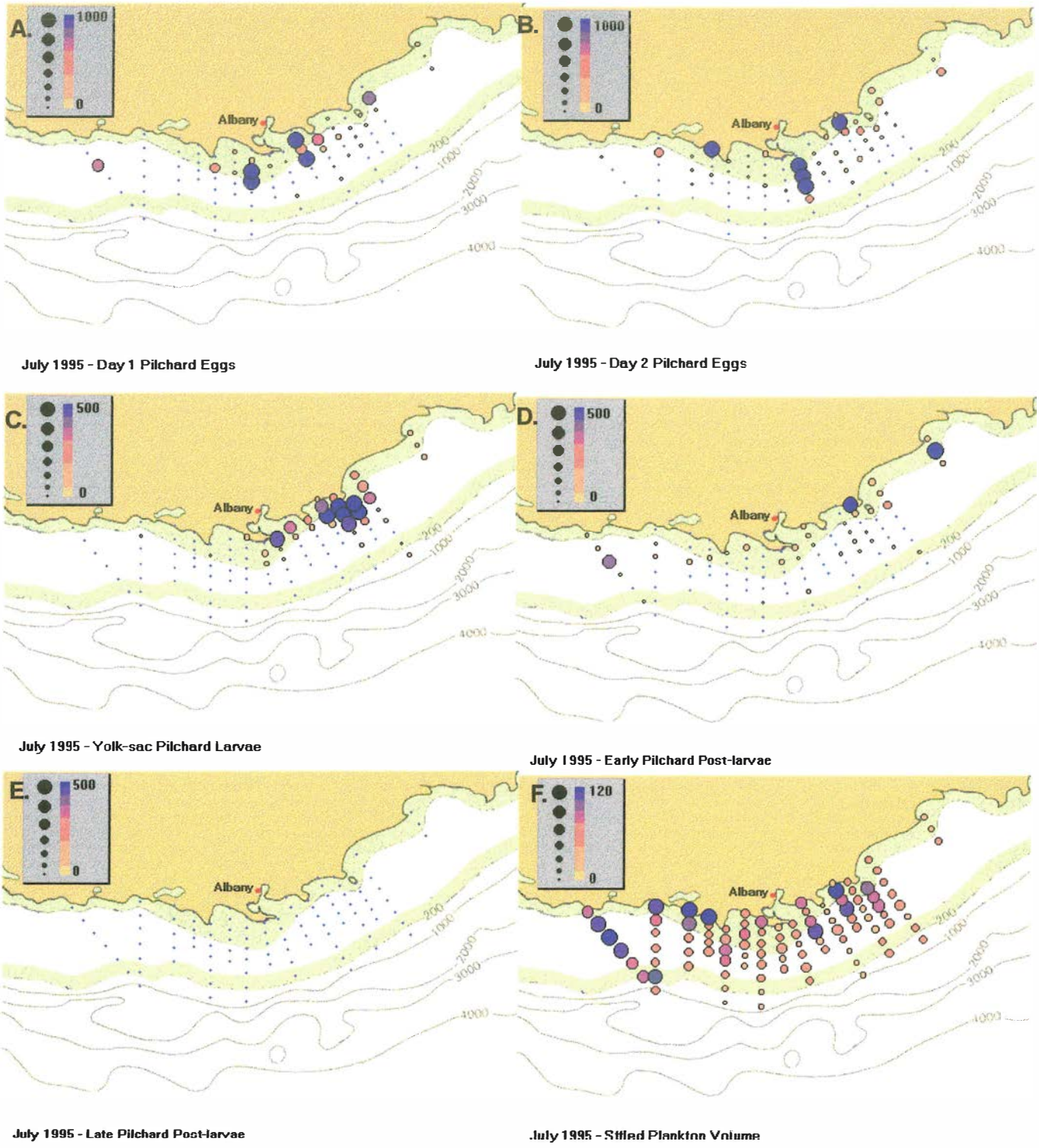


Figure 4.26: General plankton data distributions during July 1995 field trip.
A. *Day 1 pilchard eggs.* **B.** *Day 2 pilchard eggs.* **C.** *Yolk-sac pilchard larvae.*
D. *Early pilchard post-larvae.* **E.** *Late pilchard post-larvae.* **F.** *Settled plankton volume.*

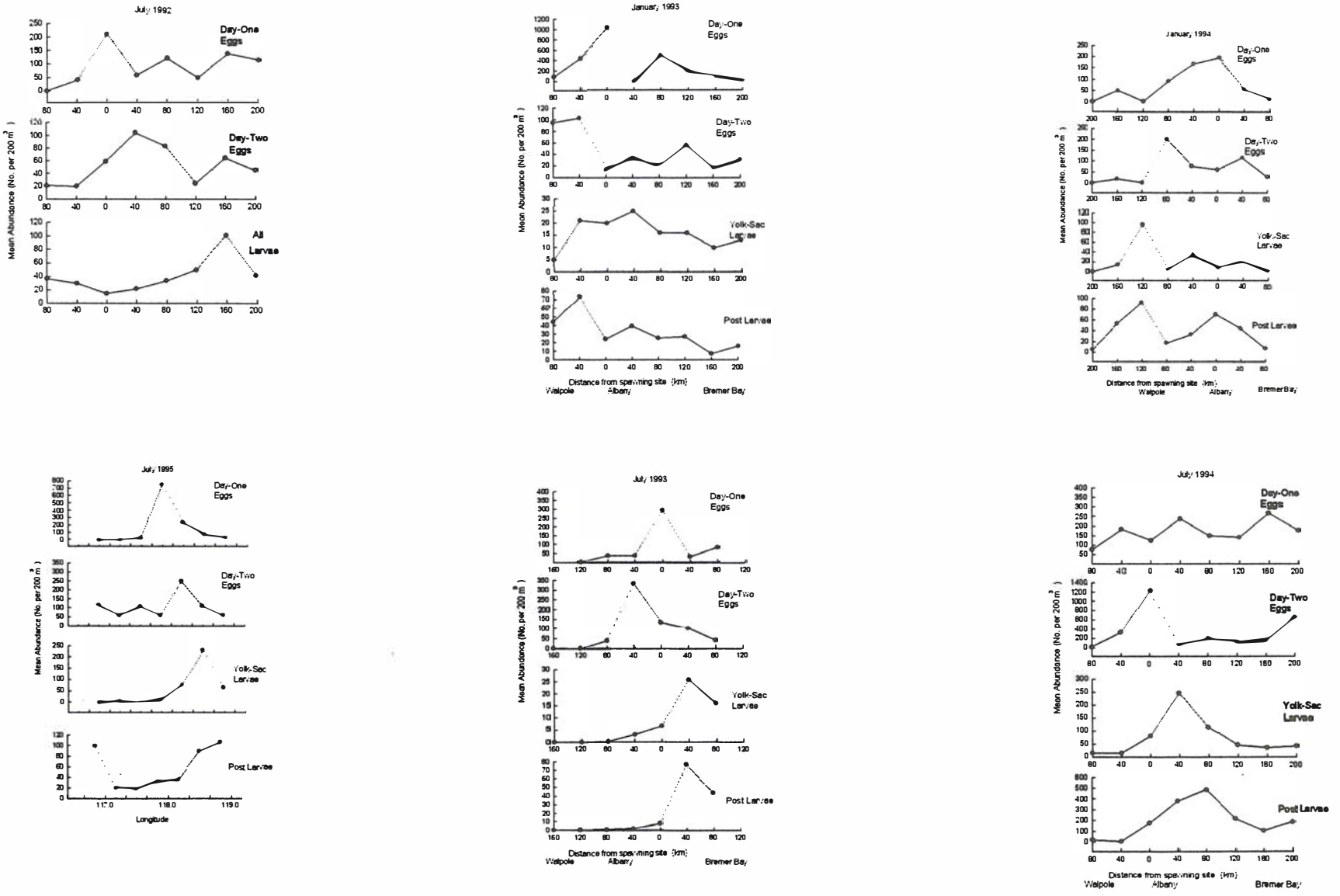


Figure 4.27 Relative distribution of pilchard stages Day-one, Day-two eggs, Yolk-sac and post larvae collected in all surveys for the region between Walpole and Bremer Bay.

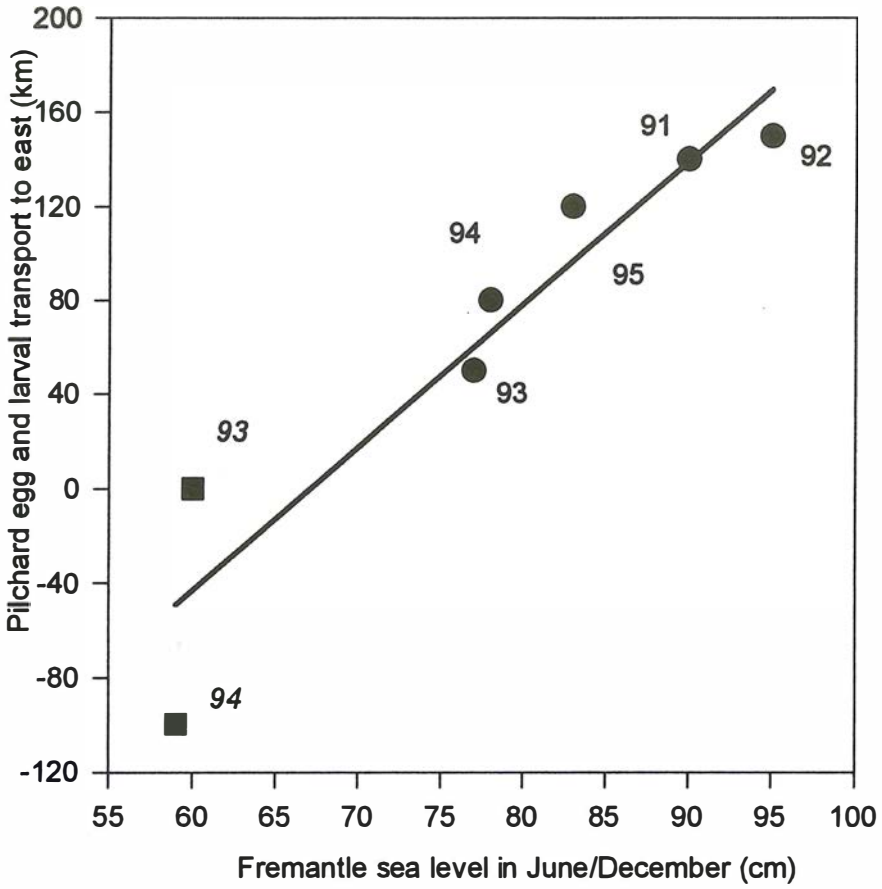


Figure 4.28: Relationship between the distance advected by early pilchard stages and the strength of the Leeuwin Current as estimated by the Fremantle sea level.

5. Section 5 Daily Egg Production Estimates of the Stock Size of pilchards in WA

5.1 A Brief History of Egg Production Methods

The concept of estimating the abundance of fish populations based upon the collection of eggs and larvae is not new, having been conceived 100 years ago by Hensen and Apstein (1897) in Norway. The procedure assumed that the number of eggs produced during a spawning season had to be a function of the mean fecundity of fish, the proportion that were female and the size of the mature stock. They considered there to be three additional assumptions for this method to work: (1) the capture of all eggs in a known volume of water; (2) uniform distribution of eggs over an extensive area; and (3) the correct identification of eggs. Unfortunately their survey failed, largely, it was reported, because the eggs that were captured could not be identified reliably.

The method was criticised later by Hjort (1912) because of the assumption that egg densities had to be uniform over an extensive area. This was probably a misinterpretation of what was meant, but there were few attempts to apply this method before the first reviews were completed by Saville (1964) and English (1964). They cited only 5 papers in which attempts had been made to assess the biomass of species that had planktonic eggs; Plaice (Buchanan-Wollastan 1923, 1926; Simpson 1959); Halibut (Van Cleve & Seymour 1953); and Californian sardines (Sette & Ahlstrom 1948).

All these studies used what is now known as the Annual Fecundity Method.

S = P /K F where:

S = Stock; P = Annual Production of Eggs, K = Sex Ratio, F = Annual Fecundity.

This method requires a knowledge of the annual fecundity of females which, at that time, was assumed to be all eggs undergoing vitellogenisis. For those with multi modes (eg sardines) there was, however, some doubt as to this calculation because it was uncertain whether there were one, two or three spawning events per year (eg Clark, 1934). The sex ratio could be calculated from catch sampling and the estimation of total annual egg production required a series of plankton tows covering the entire area and duration of spawning. The data from the series of surveys during the season is weighted up to estimate the total egg production by integrating the area under the production curve.

The process would be simplest if the spawning season was short and the area of spawning was small and defined. But as Saville (1964) pointed out, this does not fit most fishes. Furthermore, not all species have pelagic eggs, most notably, Herring (*Clupea* spp) lay demersal eggs. For Pacific herring (*Clupea pallisi*), the problem is not too great because they lay their eggs in the tidal zone which can be viewed and assessed relatively easily. Counting these eggs as a way of measuring egg production and therefore adult biomass of these stocks was first suggested by Hart & Tester (1934). The method is still in use (Schweigert, 1993).

The spawning behaviour of Atlantic herring (*Clupea harengus*) is not so accommodating; they lay their eggs on the bottom at depths not easily accessed. Runnstrom (1941) tried to sample herring eggs using a benthic grab. He obtained large numbers of samples and the counts of eggs in each grab were weighted by the area the grab represented and were then summed. Unfortunately, the biomass calculated by this method appeared to be underestimated, which he assumed occurred because some eggs would have already hatched before the sample was taken whilst others had not yet been laid.

Parrish *et al.* (1959) tried a similar method, but they first found spawning patches using a dredge and then they sampled this patch intensively. However, this method was very time consuming so they had to estimate how many patches there were.

Probably because of the difficulties sampling benthic eggs, the method was not pursued, and subsequent assessments of Atlantic herring have been attempted using surveys of larval abundances. Such estimates have also been criticised because of the level of uncertainty due to the unknown (and probably variable) rates of mortality from eggs to the larval stages that were caught (Saville, 1981). They were, nonetheless, expected to at least provide a lower limit estimate of abundance.

A second review of egg production techniques was completed by Saville (1981). By this time the situation had changed, largely as a result of fisheries management now directly affecting catch not just effort, and in some cases stopping fisheries altogether. Thus, it was becoming increasingly difficult to gain biomass estimates for many stocks using only commercial fisheries data. "In this situation, the use of fish eggs and larval surveys to estimate the size of spawning stocks is being given greater attention." Saville (1981).

Within this management climate, larval herring surveys were carried out in a coordinated and consistent basis since 1972, during which time the fishery had even been closed for periods. The variations in abundance of the larvae fitted very closely with those from VPA assessments, but it must be mentioned that the fishery data were used to scale the assessments so they were not totally independent (Saville, 1981).

According to Saville (1981), the first true fishery independent stock assessment used as the basis to set a TAC was for Atlantic mackerel (*Scomber scombrus*; Lockwood *et al.*, 1978). This was required because the fishery had expanded rapidly without catch

sampling being sufficient to provide useful information for a fishery based assessment.

A sampling program was designed to obtain an annual egg production estimate with an extensive schedule of plankton sampling completed in 1977 with 6 separate cruises covering the spawning season. Because this method assumed that all the eggs beyond a certain size will be spawned , it has also been called the total fecundity method. A number of similar programs were completed at around this time (eg Berrien *et al.*, 1981).

Concurrently, researchers in California were trying to obtain assessments for the developing northern anchovy (*Engraulis mordax*) fishery. During their intensive studies they discovered the post ovulatory follicle (*pof*; Hunter & Goldberg, 1980). This discovery, whilst useful in its own right, was made revolutionary following the suggestion by Parker (1980) that together with the measurement of other parameters a daily or instantaneous egg production method (DEPM) could be calculated which would only involve one cruise. This method relies on the percentage of spawning females being calculated (using *pofs*) to scale the concentration and area of egg production.

The discovery of pofs also negated the assumption that the number of oocytes spawned in a season were only those which were above a certain size. The number of spawning events for anchovies was originally thought to be only 2 or 3 per season, however Hunter & Goldberg (1980) found that 20-30 times per season was more likely. Thus all previous annual estimates could have been substantially wrong.

Following the testing of this method on anchovies off California, the "bible" for this technique was written by Lasker (1995). This technique involves slightly different parameters to the annual fecundity method. The parameters are:

* The area over which spawning occurred.
* The daily production of eggs (the number of eggs spawned per $0.05m^2$ per day) within the area.
* The average weight of females.
* The sex ratio.
* The batch fecundity (number of eggs spawned per batch).
* The fraction of females spawning per day.

The next major review of these methods was published following a meeting in California (Hunter and Lo, 1993). By this time there had been a plethora of egg production estimates. The DEPM had been used on anchovies and sardines in California (Bindman, 1986; Barnes *et al.*, 1992), South Africa (Armstrong *et al.*, 1988), South America (Mujica, 1986), the Mediterranean (Garcia *et al.*, 1994) and the Atlantic (Garcia *et al.*, 1991a,b). One study on Nehu (*Encrasicolina*) in Hawaii estimated biomass values smaller than 20 tonnes on a monthly basis for two years (Somerton *et al.*, 1993).

The other plankton based methods have not been rejected entirely because in many cases the daily method is not appropriate. The annual fecundity method has been used successfully on Atlantic mackerel (Lockwood *et al.*, 1981), Orange roughy (Koslow et al., 1995). Other methods still in use include a variation on this method called the fecundity reduction method (Lo et al., 1993; Zeldis, 1993), intertidal egg counts (Schweigert, 1993) and larval indices (Heath, 1993). There are also a number of comparisons with acoustic based methods (eg Armstrong *et al.*, 1988; Shelton *et al.*, 1993), which may provide better or worse assessment of stock size depending upon conditions.

The appropriate method to use will depend upon the reproduction and spawning biology of each species. It is absolutely vital to know this before you can begin to plan an assessment.

What you Need to Know

Reproduction

* Serial spawners or single batch only?
* If serial, are they batch spawners or dribble spawners. In either case, can you determine the numbers of eggs spawned each event?
* Can you determine the frequency of spawning - can you age the *pofs*, do they last 1, 2 or more days or less than a few hours?
* How long is the spawning season - a few weeks, months, an entire year?
* Do all individuals spawn on the same basis or is there some variation in timing among ages?
* Is there migration to one location to spawn with individuals moving in and out of the area or does spawning occur over the entire distribution?

Plankton

* Can you identify the eggs and how similar are they to other species in the area?
* How big is the spawning area?
* What is the patchiness of the eggs - what sampling regime do you need?
* What are the major currents?
* What is the availability of suitable vessels?

Dependent upon the answers to these questions one method may be more appropriate than another. It could also be that none of the methods are practical. This could occur from an inability to catch adults, or identify the eggs; or it may be too expensive. Nonetheless, ichthyoplanktonic based methods are now becoming widely accepted as

providing reliable estimates of spawning stock. Previously there has been some reluctance to move into this area, possibly because the techniques have been viewed as too imprecise and too expensive. The first is probably incorrect, or at least it can be determined, the second may be true for some species but it can also be incorrect.

5.2 Introduction

Most plankton surveys described in this report were specifically designed to obtain data on the spatial structure and separation of stocks along the south coast region. It was hoped, nonetheless, that the data from these samples could be used to obtain some estimates of the stock size of pilchards in the different regions. Despite only some of the surveys having been designed to obtain all adult parameter estimates required, all the plankton data are reported to indicate the level of variations possible.

5.3 Materials and Methods

5.3.1 The Model

The daily egg production method, as developed by Parker (1980), assumes that the size of the spawning biomass of a population can be estimated using the product of the daily production of eggs over the entire spawning area and the weight and proportion of females which spawned the previous night and the average number of eggs they each spawned. For continuity and ease of comparison with other similar studies we have maintained the units as defined in Parker (1985). Thus:

$$B = \frac{P A k W}{R F S}$$

where B is spawning biomass (metric tonnes), P is daily egg production (egg per 0.05 m^2 per day), A is area of spawning (km^2), W is average weight of mature females (g), R is proportion females by weight, F is the batch fecundity (number of oocytes released by an individual female of average weight), S is the spawning fraction (proportion of mature females spawning each night), and k is 20 which converts parameters from grams to tonnes and km^2 to 0.05 m^2.

5.3.2 Planktonic Egg Production

Field studies. The details of the plankton surveys are described above in Section 4. Four areas were examined separately based on the observed patterns of spawning and additional information on stock separation; Albany (Longitude 117° to 119° E), West Coast (Latitude 31° to 33° S), Bremer Bay (Longitude 119° to 121° E), Esperance (Longitude 121° to 124° E).

Laboratory analysis. From the samples collected, we classified the eggs from each cruise as either having been spawned the previous night (8-16 h old, Day-one) or two nights previously (28-40 h old, Day-two). In some surveys, the eggs were classified into four age categories, those between 3 and 12 h, 12 and 18 h, 33-39 h and 39-42 h (see Section 2 for stages).

Egg production estimation. The total spawning area (A) of pilchards was defined as the area in which Day-one pilchard eggs were found. This was calculated by delineating the areas containing this egg stage, but this often included a few embedded zeros. The production of eggs (P) from the bongo net samples were calculated by assuming that the region between 50 m and the surface was sampled in a uniform manner for the vertical tows and once the oblique tow data were transformed by the conversion ratios above. The abundance index for each tow (both vertical and oblique) was converted using General Oceanics flowmeter readings to obtain the rate of

production per 2.5 m^3 (which equates to 0.05 m^2 for a depth of 50m). For the 1995 surveys, which used the CalVET nets, the data were used assuming each net sampled 0.05m^2.

Egg production, P_O, was estimated by calculating the average abundance of both Day-one and Day-two eggs. Where sufficient data was obtained, an exponential mortality function was fitted to the abundance of eggs for each half day interval (0600-1200; 1200 -1800) using the formula:

$$P_t = P_O \, e^{-zt}$$

where P_t is the abundance per 0.05 m^2 of eggs in age category t, P_O is the daily egg production per 0.05 m^2, t is the estimated time in days elapsed since spawning, and z is the instantaneous rate of daily mortality.

5.3.3 Adult Weight, Spawning Fraction and Fecundity

Field studies. Samples of adult sardines were obtained from the local purse seine fleets whenever possible. The numbers obtained varied from 0 - 300 individuals per survey depending upon availability.

The gonads of the female fish obtained were fixed in 10% buffered formaldehyde. These gonads were subsequently transferred to 70% alcohol and prepared for histological analysis. Additional female gonads, which were visually staged as ripe (gonad stage 4; Matthews 1964), were collected and kept in 10% formaldehyde for the determination of their batch fecundity.

5.3.4 Laboratory studies.

Spawning fraction. Thin transverse sections of the female pilchard gonads were cut, mounted on slides and stained using standard haematoxylin and eosin solutions. Each of these slides were examined for the presence of Day-0 (8-23 hrs old; Fig. 5.1a) and Day-1 (23-48 hrs; Fig. 5.1b) post-ovulatory follicles (*pofs*) as shown for *Sardinops sagax* from Peru (Goldberg *et al.* 1983; Hunter and Macewicz 1985). The proportion of females with Day-0 *pofs* were used to estimate the spawning fraction.

Batch fecundity A standard gravimetric method of calculating fecundity was used (eg. Le Clus 1977; Hunter *et al.* 1985) whereby the gonads of each suitable fish (those with stage 4 a,b,c gonads; Fig. 5.1c,d,e) were weighed and three sub-sections were removed, weighed (each of approximately 0.01-0.03 g), teased apart on separate slides and the number of mature oocytes counted. Discrimination of the mature oocytes was facilitated by there being three readily identifiable stages of oocytes in ripe female gonads (Clark 1934; Le Clus 1977). Only the most advanced oocytes were counted. It has been found (Fletcher unpublished data) that fecundity estimates for WA pilchards using ovaries with ripe gonads were not significantly different to those with hydrated oocytes and were therefore suitable for use.

The batch fecundity for each individual was calculated by the product of the mean number of mature oocytes per gram of gonad and the total gonad weight. A linear regression of batch fecundity against gonad-free weight for all the females examined was then calculated. If no samples were available, mean values for the site and the time of year were used (Fletcher unpublished).

Parameter Estimation The means and variances for each of the parameters (W, R and S) were calculated from the sample data weighted by the number of individuals within each of the samples (Picquelle and Stauffer 1985). For F, the linear regression of

batch fecundity against ovary free weight was used to provide an estimate of the batch fecundity for every mature female in each sub-sample. These estimates were used to calculate the average batch fecundity for the entire survey with the variance about the slope of the regression line used as part of the calculation of the variance about this mean (see Picquelle & Stauffer, 1985 page 10). It was assumed that the fish obtained from the commercial catches used here provided an unbiased sample for the estimation of the weight, sex ratio and fecundity of the spawning population in the region.

Coefficients of variation (CV) were calculated for each of the parameters and estimates of the covariance (COV) were calculated for each pair of parameters (see Parker 1985).

5.4 Results

5.4.1 Albany (Table 5.1)

In the Albany region the total spawning area of pilchards varied between surveys from 3000-5600 km^2 . This comprised between 25% and 53% of the total area surveyed (Fig 5.2). In general total spawning area was greater in summer surveys than winter surveys, however the greatest spawning area was attained in July 1994, a time when modeling calculated the stock size was the largest.

The density of pilchard eggs within the spawning area also varied. Values for Day-one eggs varied from 1.4 - 7.1 per 0.05m^2 and for Day-two eggs, values between 0.91 and 7.2 were obtained. In three surveys (July 92, January 93 and July 95) the numbers of Day-one eggs exceeded that of Day-two eggs, which allowed a detailed assessment of P_0 using the egg mortality model. In the other three surveys, however, Day-two eggs were the most common stage present. The P_0 values used varied from 2.3 in July 1993 to 9.9 in July 1995.

Spawning frequency was estimated in 4 of the 6 surveys. These, like egg production, varied greatly from 4.3% in July 1993 to 18.8% in July 1995.

Adult weight did not vary substantially among surveys except for January 1994 where large individuals were collected. The average fecundity during winter surveys were similar at approximately 11000, but summer surveys had values > 15000, reflecting the better condition values found in this season (Fletcher unpub).

The calculated biomass values were reasonably similar. The lowest values were recorded in July 1992 and 1995 at approximately 17000 t, and the highest value in July 1994 which was in excess of 30000 t. The CV values for the 3 surveys where all parameters were estimated were between 0.22 and 0.42. The maximum and minimum biomass values reported were calculated by using the largest and the smallest possible value for each parameter. These should give a range in which the true biomass is most likely located.

5.4.2 Bremer Bay (Table 5.2)

Surveys in this region were hampered by a series of breakdowns to the vessels chartered to complete this region resulting in the area not being fully surveyed on two occasions (see Section 4). Thus only in July 1992 and 1994 was the area sampled on a realistic basis (Fig. 5.3). Furthermore, the lack of resources meant that all adult parameters were obtained from this location on only one occasion.

The spawning area was reasonably large at > 5000 km^2. The numbers of eggs collected for both categories were moderate with averages between 2.1 and 4.8 per 0.05m^2. These resulted in P_0 values of 3.5 - 6.5.

Adult weight and batch fecundity values in the years when they were measured were very similar at approximately 38 g and 12000 respectively. The only survey in which spawning frequency was estimated was July 1993 where a relatively low value (2.5%.) was found.

Spawning biomass values between 20000-40000 tonnes were calculated for this region. Whilst the precision of these estimates is not great, it is noteworthy that the minimum estimates were all above 10000 t.

5.4.3 Esperance (Table 5.2)

The area of spawning at Esperance was relatively large at 7000-9000 km^2 (Fig. 5.3) The surveys conducted in July collected relatively few eggs with P_0 values of 0.7-2.5. In the May survey, however, reasonable numbers were collected with a P_0 of 6.

Individuals, and therefore batch fecundity, tended to be larger in Esperance. The single estimate of spawning frequency obtained from this site in July 1993 was relatively low at 2.3%. The values for July 1994 and May 1995 were assumed to be higher since greater numbers of eggs were found. An unfortunate consequence of the pilchard mortality event was the cessation of fishing in Esperance during the period that the plankton surveys were conducted which precluded obtaining a complete set of parameter estimates.

The calculated biomass values for this region indicate a relatively large stock, possibly in excess of 30000 t even when the relatively large, and therefore conservative, values for spawning frequency were used. Like Bremer Bay, all the minimum estimates were greater than 10000 t.

5.4.4 West Coast (Table 5.3)

Sampling on the west coast was usually an adjunct to the south coast work and hence not as thorough (Fig. 5.4). Adding to this is the increased shelf width present in this area which greatly expands the area that needs to be sampled. The areas sampled by the surveys in this region varied from 9 000-16000 km^2. Similarly the measured spawning area also varied from 6 000-12000 km^2.

The density of pilchards eggs was very large in July 1993. The P_0 value used of 10 per 0.05 m^2 per day was less than was originally calculated (15). One plankton tow collected over 3000 eggs (the largest for the entire study) which doubled the average catch of the survey. If this sample is ignored P_0 would have only have been 7.

Unfortunately we have been unsuccessful in getting any adult samples during the times that the plankton surveys have been completed. Thus all adult parameters have been assumed from other work completed in the area. For example adult weight off the west coast are significantly greater than for the south coast (Fletcher unpub).

Utilising these assumed values, the calculated biomass levels have varied from 8000 to 40 000t. Obviously, with the difficulties in the parameter estimates used, little confidence can be placed in these values. It is noteworthy that the minimum values vary from 3000 to 14 000 t.

5.5 Discussion

The application of the DEPM has been very useful for the management of pilchard fisheries in WA. A total of 7 estimates of biomass for the Albany region have been calculated, 4 of which have all parameters estimated. In all of these, the calculated value has been between 16000 and 32000 tonnes. The CV values have been between

0.29 and 0.44 which are within the range found in other studies (Lo, in press). These data support the other information collected on the stock size for this region in that the biomass is probably less than 30000 t in most years.

The variations in biomass estimated using the DEPM were reasonably consistent (r = 0.72, P = 0.06, n = 7) with the equivalent biomass values estimated by modelling (Fletcher, 1992, 1995). For the July surveys, the relationship between model generated biomass values and spawning area, whilst not significant, was positive (r = 0.5, P = 0.3, n = 5) whilst the relationship between the DEPM biomass and spawning area was highly significant (r= 0.91, P = 0.03, n = 5).

The estimates of biomass obtained using the DEPM for the other 3 sites were not as reliable because the same level of effort could not be expended with the resources available. Nonetheless, these preliminary data have shown that there are probably considerable stocks of pilchards located off Bremer Bay and particularly Esperance. The biomass estimates for Bremer were all above 15000 t and for Esperance values approaching 30000 were calculated. The vast spawning area of pilchards off Esperance makes it unlikely that there is only a small stock in this location. These data have now been used to assist with the recent allocation of a TAC for the Esperance area. The initial quota was set at 1200 t. This has now been raised to 1800 t largely on the basis of the plankton data and will be reviewed again shortly.

The values for the West Coast are the least reliable. In no survey were any of the adult parameters available which make the calculated values most tenuous. Thus these data have not been used to make any management decisions concerning the fishery in the west coast. A dedicated effort to sample this region is required and is planned for the next two years such that the quota for the region can be determined.

It has been decided that in future, with the limited resources available, sampling needs to be restricted to only one of the regions per survey period. This will allow a much greater sampling effort in the one zone and therefore improve both the precision levels of the estimates obtained and the chances of obtaining estimates of all parameters.

The last problem to be determined during this project was the problems with using large plankton nets to conduct the vertical tows. These nets are affected greatly by windage such that in rough weather they become semi-oblique tows, this probably contributed to the odd egg production results in some surveys. The recent use of the CalVET appears to have reduced this problem. They are easier to keep vertical even in very windy conditions. The potential reduction in the measured spawning area (see section 8), did not appear to be evident in the May 1995 Esperance survey. But this could be a problem if sampling was completed during a time when spawning frequency, and hence egg production, was low.

	July 92	Jan 93	July 93	Jan 94	July 94	July 95
Area sampled (km^2)	8856	10210	11620	10590	14360	12030
Spawning area (km^2)	3788	5466	3923	4850	5625	3079
Number of tows	41	43	85	55	74	100
Number of tows in spawning area	30	31	41	34	52	50
Day-one eggs	2.9	7.1	1.4	2.7	2.7	6.7
Day-two eggs	1.6	0.91	2	3.9	7.2	1
Ave	2.2	4	1.8	3.3	4	4
P_0 value used	3.9	10.3	2.3	4	7	9.96
Adult Wt	34.6	36.8	34.1	46	36.6	34.6
Batch fecundity	10950	17800	9834	15000	11000	11200
No. used for fecundity	18	43	34	0	5	45
Calculated spawning freq	9%		4.3%		13.8%	18.8%
Number of histo sections	70	78	171	128	138	300
Assumed S.F.		15%		10%		
Biomass	16994	25530	23432	20615	31330	17600
CV	0.44		0.299			0.21
Min estimate	9000	11000	9000	12000	15440	8000
Max estimate	25000	66923	45000	40000	55000	28000

Table 5.1: Albany Region DEPM Estimates. Shaded boxes indicate that these were assumed values.

	Bremer Bay			Esperance		
	July 92	July 93	July 94	July 93	July 94	May 95
Area sampled (km^2)	9085	5488*	10608	17285	17660	12000
Spawning area (km^2)	5313	3108*	5947	7914	7303	9700
Number of tows	29	27	44	54	18	110
Number of tows in spawning area	21	16	29	26	14	100
Day-one eggs	3.6	2.93	5.38	0.66	2.03	4.75
Day-two eggs	3.8	1.29	4.25	0.52	1.86	1.65
Ave	3.7	2.1	4.8	0.6	1.9	3.2
P_0 value used	4.0	3.5	6.5	0.7	2.5	6.0
Adult Wt	39.6	36.3	39.6	47.5	39.8	42
Batch fecundity	12862	11631	12000	11820	12000	17000
No. used for fecundity	19	31		12		
Calculated spawning freq		2.5%		2.3		
Number of histo sections		101		80		
Assumed S.F.	10%		15%		10%	15%
Biomass	19280	44010*	28458	32252	20080	31900
Min estimate	12000	11370*	15700	14200	10700	10900
Max estimate	79000	74000	42500	61800	40100	45647

Table 5.2: Bremer Bay and Esperance Regions DEPM Estimates. * indicates less than full area of stock covered. Shaded boxes indicate that these were assumed values

	July 93	Jan 94	July 94
Area sampled (km^2)	16356	13434	9390
Spawning area (km^2)	12500	6558	5775
Number of tows	37	35	51
Number of tows in spawning area	30	20	25
Day-one eggs	13 (5)	2.71	1.14
Day-two eggs	1.1	3.86	.41
Ave	7.2 (3.3)	3.3	0.75
P_0 value used	10	4	2.1
Adult Wt	41.6	41.59	38.8
Batch fecundity	20000	20000	18000
No. used for fecundity			
Calculated spawning freq			
Number of histo sections			
Assumed S.F.	20%	10%	10%
Biomass	41250	18362	8714
Min estimate	14500	4900	3100
Max estimate	78000	36700	29000

Table 5.3: West Coast DEPM Estimates

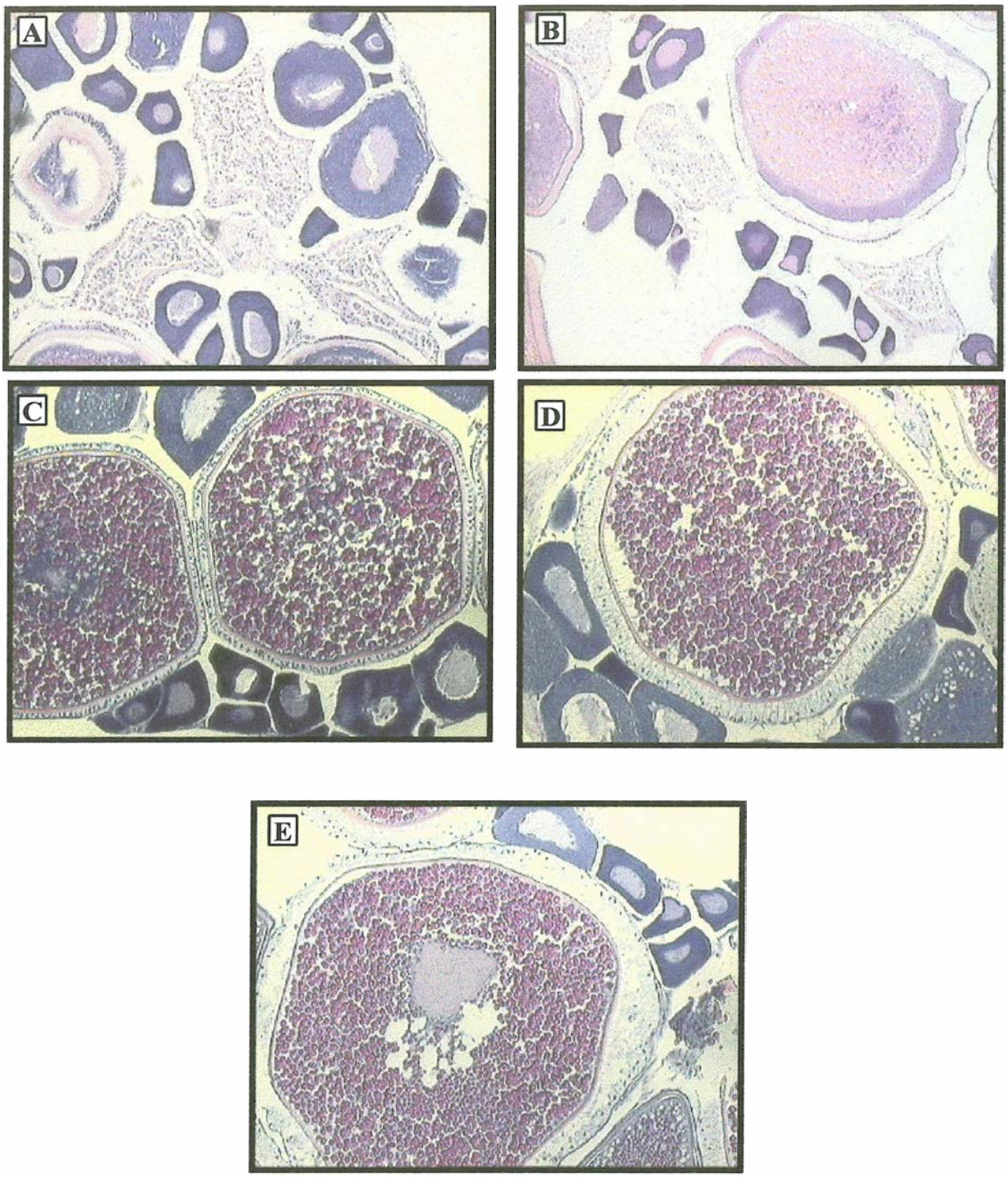


Fig. 5.1: Pilchard post-ovulatory follicles. (a) *Day-zero (stage 6a)* (b) *Day-one (stage 6b)* ; and ripe gonad stages (c) *ripe (stage 4a)* (d) *columnar epithelium (stage 4b)* (e) *migratory nucleus (stage 4c)*

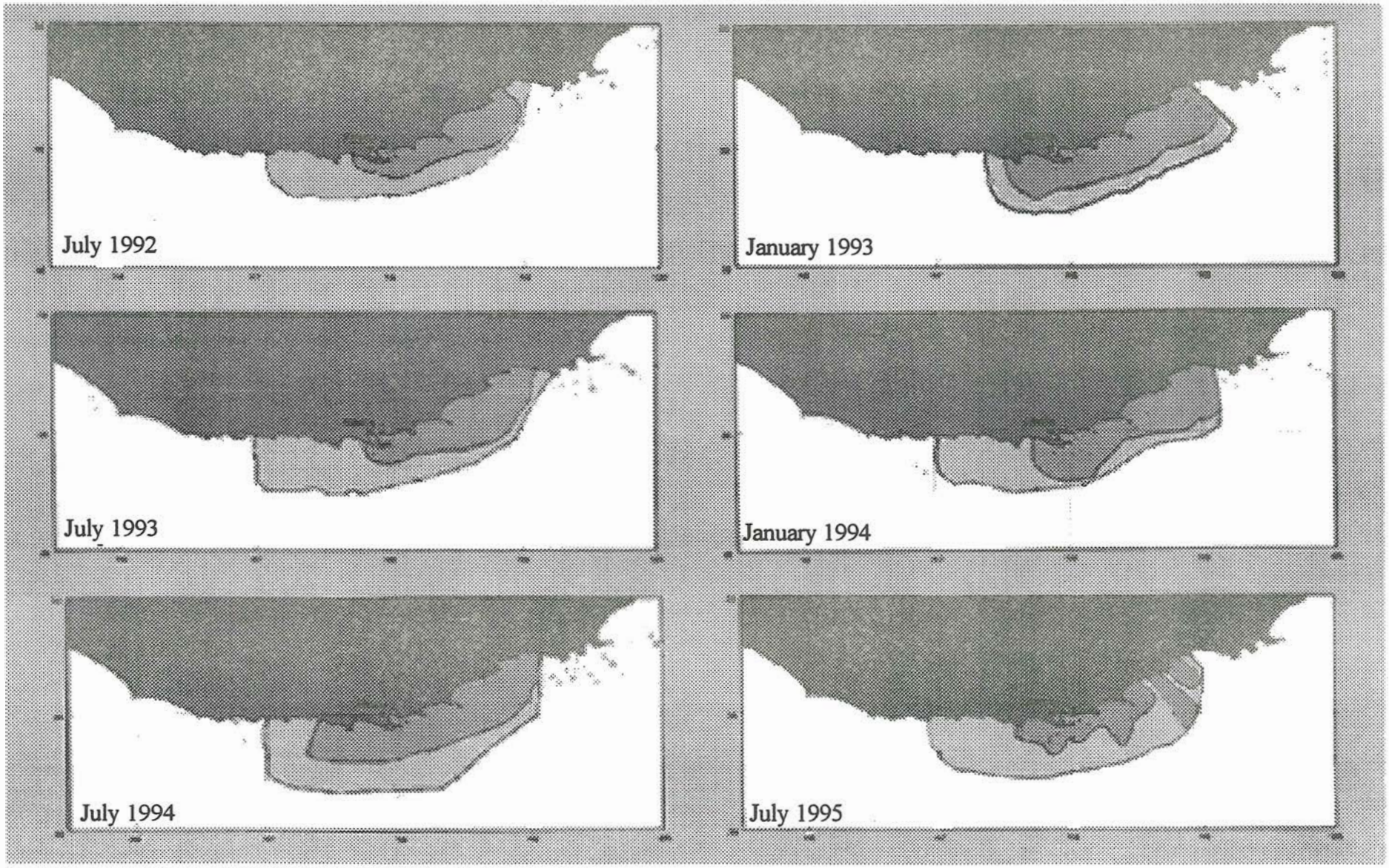


Fig. 5.2: Spawning areas and survey areas of the Albany region.

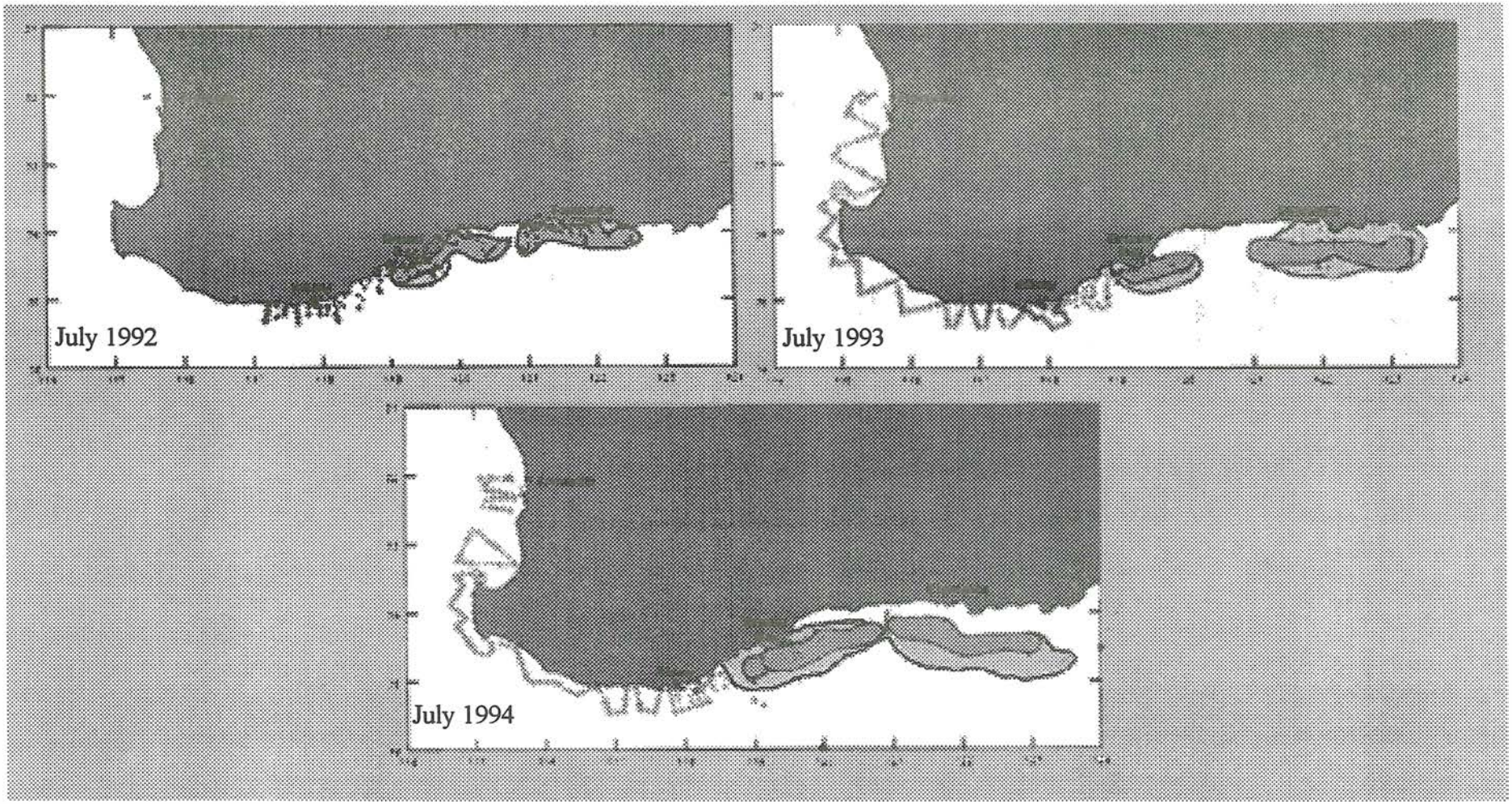


Fig. 5.3: Spawning areas ▒ and survey regions □ for the south coast showing the Bremer Bay and Esperance regions

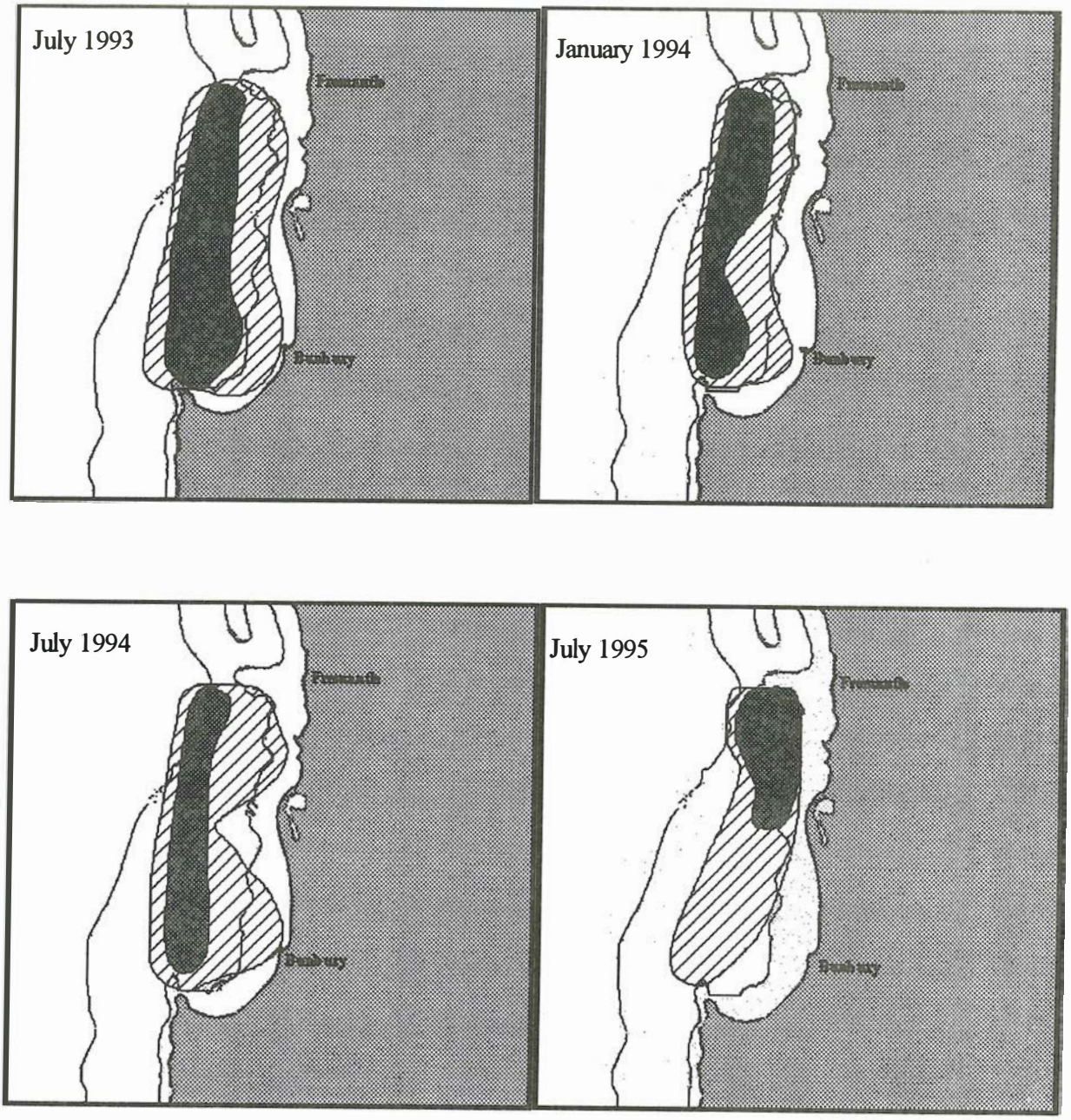


Figure 5.4: Spawning areas ▓ and survey regions ▨ for the west coast

6. Section 6: Spatial distribution of pilchard eggs and larvae as determined from close observation sampling.

6.1 Introduction

The spatial distribution of organisms has a large impact on the most appropriate sampling scheme for accurately estimating their abundance. This knowledge is especially necessary if these samples are used to compute estimates for stock assessment purposes. Fisheries scientists have been concerned with the spatial variation of samples they obtain from the oceans for many years. However, it is only recently that satisfactory means have become available for describing the observed variation quantitatively through the study of geostatistics.

Geostatistical techniques originated in the mining industry for the analysis of ore samples (Matheron, 1965, 1971). Their use within marine science area can be used for the design of optimal sampling schemes and the spatial prediction of abundances. A number of fishery resource assessment techniques rely upon spatially separated samples. For example, the daily egg production method (DEPM) depends upon using the abundance of eggs collected by numerous plankton hauls to estimate the extent and density of eggs over large areas (see Section 5 for details). However, plankton studies often produce highly variable data due to the very patchy distributions of eggs and larvae. Hence prior knowledge of the nature and scale of this variability will help design the most appropriate survey.

Only a few studies have estimated the spatial distribution of fish eggs collected by plankton tows. Most of these, however, examined only relatively small scale variability by collecting samples at the same locations a number of times (eg Silliman,

1946; Smith & Hewitt, 1985). None have directly determined what is the most efficient sampling distance among stations for the sardine *Sardinops sagax* for the completion of a DEPM survey. In this study we use data obtained from plankton tows which were conducted on a fine scale grid basis to investigate the variability in abundance of pilchard eggs and larvae over the range from a few centimetres to thousands of metres.

The data will be analysed using variograms which spatially represent the levels of relatedness of samples at increasing distances apart. A correlogram will also be used to determine the level of autocorrelation among samples at varying distances apart. This will be the first time these techniques will have, to our knowledge, been used on plankton data. Finally, jackknifing will be used to examine the potential variation in abundance that results from a number of different sampling intensities. From these analyses, the preferred distances apart for future surveys will be determined.

6.2 Methods

6.2.1 Plankton Sampling

A large number of vertical plankton tows were completed within a relatively small regions of the south coast near Albany in both July 1993 and January 1994 (Fig. 6.1). The general survey methods were similar in both surveys with stations completed on a grid basis with varying intervening distances. Thus, stations were either 1, 2 or 4 nm apart. Furthermore, more than one plankton haul was often made at the same station to investigate variations at the scale of a few metres. Finally, at most sites, particularly in January 1994, each bongo net on the frame was analysed separately to gauge the fine scale (cm) variations in abundance.

In July 1993 a single vessel completed the tows using a satellite derived Global Positioning System (GPS) to determine station locations. Sampling occurred from 0600 to 1800 hours. In January 1994 two GPS equipped vessels were used simultaneously between 0700 and 1500 hours. The nets used were 600 mm diameter standard Bongo nets, with both having 500 μm mesh or with one having a 300 μm mesh, the other had 500 μm mesh. The different mesh sizes were used to assess possible mesh selectivity in which it was shown (Section 8) that there is no difference in the abundance of eggs retained by the two mesh sizes. For larvae, however, only the 300 μm mesh samples were analysed since substantial extrusion of early stage larvae occurred in the coarser mesh net (see Section 8). At the sites where two hauls were completed, two separate sampling units were used to avoid any potential contamination. Where each net was assessed separately, a General Oceanics flowmeter was located in the mouth of each net. For tows where the samples were combined, only a single flowmeter in one net was used.

The samples were sorted and staged according to the general methods detailed above (see Section 2) with the resultant data analysed using the relative indices of abundance per 200 m^3 calculated for all pilchard eggs, Day-one and Day-two eggs and the three larval stages (Yolk sac, early and late post larvae).

6.3 Results

The indices of abundance for pilchard eggs collected in the two surveys are shown in Fig. 6.1. In general, the first survey found few eggs and larvae with only one patch of eggs. The overall abundance was low with the average abundances of pilchard eggs and larvae were only 42 and 6 per 200 m^3 respectively. In the second survey, large

numbers of eggs and larvae were collected with mean abundances of 360 and 573 respectively. Consequently, the latter survey could be analysed more thoroughly.

6.3.1 Detecting the Presence of Spatial Structure

The nature of spatial structure (autocorrelation) between samples is of interest. It is useful to know that if a first sample (A) is drawn and recorded, then if another sample (B) is taken near the first, can the approximate value of a particular variable at (B) be estimated. This is determined by the existence and nature of spatial structure. If the two values are not independent, the value of the variable at (B) will be related to the value of the same variable at (A) and the distance between (A) and (B) is within the zone of spatial influence of the underlying ecological phenomenon. Autocorrelation often depends on the distance between the two sampling points with positive values corresponding to short distances and zero or even negative values corresponding to large distances.

6.3.2 The Correlogram

Spatial structure can be represented as a graph where the autocorrelation is plotted against distance. This is called a correlogram and the distance may be in one direction or, in the case of the all-directional correlogram , all directions. The all-directional correlogram and omnidirectional variogram (see below) make the assumption that the spatial structure is isotropic. This means that the autocorrelation is the same regardless of the direction being considered. When the autocorrelation is a function of both

distance and direction the structure is anisotropic and should be looked at in one direction at a time.

The spatial structure of the abundance of Day-one pilchard eggs in the January 1993 was investigated using a two-dimensional correlogram as shown in Fig. 6.2. The correlogram shows the spatial structure as a function of distance with contours depicting the autocorrelation values. The resultant pattern is symmetrical since the autocorrelation is the same regardless of whether one is travelling from east to west, or west to east. This applies for all opposite directions. The zone of spatial influence (taken as the distance from the centre of the graph to the zero contour line) is slightly greater in the east-west direction, which largely follows along the coastline, than the north-south direction which is largely the inshore-offshore direction.

The area of spatial influence corresponds to the distance from the centre of the patch of eggs to the edge of the patch in a given direction. Since this represents each single patch of eggs, the distance from the zero contour across the centre of the figure to the opposite zero contour corresponds approximately to the average size of patches in that direction. Using this technique, the average size of the patches of Day-one eggs during January 1994 was estimated to be 0.13 degrees (7.8 nm) across in the north-south direction and 0.17 degrees (10 nm) across in the east-west direction.

6.3.3 The Sample Variogram

The variogram is fundamental to geostatistics. It describes spatial variation quantitatively in terms of distance and direction, and in a way that facilitates our understanding of spatial patterns and processes. The variogram is also central to techniques for spatial prediction such as kriging (Krige, 1966). Given values for a variable Z_t are the realisation of a random function $\{Z_t; t \in R^2\}$ observed at certain points $\{t_i\}$ of a region R^2. The variogram is a function of distance and can be estimated from the data Z_{t_i}, i=1,...,n as follows:

$$\hat{\gamma}(h) = \frac{1}{2N_h} \sum_{i=1}^{N_h} (Z_{t_i+h} - Z_{t_i})^2 \qquad h = h(1), h(2), \ldots, h(k) \tag{1}$$

where $h(1)$, $h(2)$,...,h(k) are discrete lags and N_h is the number of lag-h differences. Before using the variogram one must assume that the data are stationary (the mean and variance of the data are constant over the area of study), otherwise trends (ie. drift) in the data will need to be removed (Cressie, 1991). The variogram values lags and number of lag-h differences for the omnidirectional variograms of Day-one and Day-two pilchard eggs at two locations in the southern ocean was computed using GEO-EAS (Englund and Sparks, 1991). We assumed that the spatial structure is isotropic and used the omnidirectional variogram. Although directional variograms for one of the locations indicated that the range was slightly longer in the east-west direction than the north-south direction. The omnidirectional variogram contains more sample pairs than directional variograms, and is, therefore is more likely to show a clearer spatial structure.

6.3.4 Modeling the Sample Variogram

Many geostatistical studies require estimation of the variogram from the data. A theoretical variogram (model) is fitted to the experimental variogram so the variogram values may be readily estimated for any distance within the range of the data.

The important features of the variogram to be estimated are:

Range (a): A measure of the size of spatial influence of the process, which is the distance at which the variance stops increasing and the sill is achieved. At distances greater than the range an increase in the separation distance between pairs of sample points no longer results in an increase in the variance.

Sill (C): The value for $\gamma(h)$ at the plateau that the variogram reaches when the variance is no longer an increasing function of distance.

Nugget Effect (C_0): The distance between the origin and the value of the variogram at extremely small separation distances. This effect is due to sampling error and small scale heterogeneity which often produce a value greater than zero.

When used for kriging, functions that model the variogram must produce a covariance matrix that is positive semi-definite. This ensures that predicted random variables have a positive variance and that a unique solution to the kriging equations exists. The variogram may be fitted by one or a combination of several variogram models by estimating $\theta = [a, C_0, C]^T$. The spherical exponential and Gaussian models (Isaaks

and Srivastava, 1989) were investigated using a Mathcad (Mathsoft, 1995) worksheet developed by the authors. Weighted least squares as suggested by Cressie (1985, 1991) was used to fit a model to the experimental variogram by finding the value of θ that minimises Eqn(2)

$$S(\theta) = \sum_{j=1}^{k} \frac{N_{h(j)}}{(\gamma(h(j);\theta))^2} \{\gamma^*(h(j)) - \gamma(h(j);\theta)\}^2 \qquad (2)$$

where $g^*(h(j)); j=1,..., k$ are the variogram values and $g(h(j)); j=1,..., k$ the modelled variogram. The objective function places more emphasis on the small lags with a larger number of pairs near the origin. This is desirable since when the variogram model is used for kriging the fit near the origin is more important than the fit at larger lags. The spherical model (Eqn(3)) was selected since it gave the best fit with the lowest sum of squares value.

$$\gamma(h;\theta) = \begin{cases} C_0 + C\left(\frac{3h}{2a} - \left(\frac{h^3}{2a^3}\right)\right) & h < a \\ C_0 + C & h \geq a \end{cases} \qquad (3)$$

The variograms of the egg and larval stages are shown in Fig. 6.3. These show the isotropic structure of the patches following spawning. Samples (tows) that are spatially close were similar, with an associated small variance, while samples that were further apart were less similar and consequently had a larger variance. The variance increases with increasing distance between a pair of sample points until the sill is achieved. The maximum variance in a patch will be attained at a distance equal to the radius of an aggregation since at this distance high abundance samples in the centre of the aggregation are being paired with low abundance samples on the edge. As the lag is increased further, the variance decreases as sample pairs on opposite sides of the aggregation with similar low values for abundance predominate. At a lag

equivalent to the diameter of the aggregation or larger the variance is small since both samples in any pair are likely to miss the aggregation and have similar low values for abundance. The sampling interval should be smaller than the diameter of the patch to ensure that the patches are not overlooked and contain at least one sample. If the sampling interval is too large compared to patch size, entire patches may be overlooked.

The distance where the sill value was achieved for Day-one eggs was at 0.12 degrees (7 nm) with values approaching the minimum again by 0.3 degrees (20 nm; Fig. 6.3a). For Day-two eggs the fitted curve had a calculated sill value of 0.04 degrees (2.4 nm) but the largest value of variance was attained at a distance of 0.13 degrees (7.8 nm) with small values again attained by 0.3 degrees (20 nm; Fig. 6.3).

The data for Day-one eggs were examined separately for variation in the east-west and north-south directions (Fig. 6.3 c,d). In the east-west comparison, the sill value was close to 0.18 (11 nm), whilst in the north south direction it was only 0.1 (6 nm).

Yolk sac larvae also had a large degree of spatial cohesion with increasing levels of variance in sites up to 0.13 degrees (8 nm apart; Fig. 6.3.c). For both of the post-larval stages, however, the spatial structure had altered such that there was a high level of variance even in close samples. Thus no curve was fitted (Fig. 6.3d,e).

In the July 1993 survey, in which few eggs were collected, there was still a discernible spatial pattern (Fig. 6.3f). The distance at which the sill was achieved was relatively large at 0.35 degrees (21 nm). The large increase in the variance after this distance is an artefact of the single large value present in this survey.

6.3.5 Spatial Prediction

The kriging method is used to compute an estimate for a point from surrounding observations within the range of spatial influence. The observations are weighted using the distance-variance relationship from the variogram. Kriging is often used to generate a three dimensional mesh of x-y coordinates and predicted values, which, when displayed as contours or a three dimensional surface, enhances our understanding of any spatial relationships in the data. Ordinary kriging was used to estimate the abundance of one and two day old pilchard eggs in the study area as shown (Fig. 6.4a, b). The distribution of eggs is obviously not homogenous but rather clustered in patches. We cannot assume that the population is distributed either uniformly or randomly in space. This has implications for selecting an appropriate sampling strategy that takes account of the heterogenous spatial distribution.

6.3.6 Jackknifing

The data for the January 1994 survey were subjected to a jackknifing analysis. The region was divided into 6 sub-areas, each of which had between 6 and 9 representative tows. A total of one, two or three tows were selected randomly (without replacement) from each of these 6 areas and combined to calculate an estimate of the population mean value. This sampling was completed a total of 1000 times.

The population mean for Day-one eggs (using only one sample per site) for all 48 sites was 96. However, the distribution of means calculated using 1 sample per zone (6 in total) varied from 15 to 450 (Fig. 6.5). The majority (90%) were, nonetheless, between 25 and 150. The bimodal nature of the distribution was from the impact of

one large value being either present or absent from the random samples. By increasing number of samples to calculate the mean, the total range of means declined but the range for 90% limits did not decrease greatly with 2 or even 3 samples per zone (Fig. 6.5).

6.3.7 Bootstrapping

Both the raw and kriged data were subjected to a bootstrapping exercise. Random samples of between 2 and 100 selections were made 1000 times with the mean values calculated. The standard deviation of these 1000 means was large at small sample sizes but dropped rapidly to a plateau beyond 20 samples (Fig. 6.6). Similarly, the 90% confidence limits on the kriged data declined from a range of 50-500 to 100-350 when only 2 samples are taken in this size region compared to when 12 samples are taken. Further increases beyond 24 samples yielded little further reduction in these limits (Fig. 6.6).

6.4 Discussion

The data we collected here support the notion that fish eggs are not distributed randomly throughout the oceans. We found a large degree of concordance between the numbers of pilchard eggs and early larval stages collected by replicate tows at the same sites. This endorses the concept of using plankton sampling as an accurate, quantitative method. The repeatability of samples taken at the same location in quick succession has also been investigated by Silliman (1946) who, from replicate tows at a number of sites, found that 90% of the variability in abundances of *Sardinops* eggs off California was due to differences among sites with only 10% due to sampling variability. Similarly, Smith & Hewitt (1985) found that there were strong similarities in the numbers of Day-one to day 3 anchovy eggs within the 8 samples they took at

each site. They concluded that nearly uniform results would have been obtained irrespective of which of the replicates had been used.

The number of spatial scales sampled in this study allowed the determination of the area over which spatial cohesion exists. The level of correlation in abundance among sites declined steadily with increasing distance for most stages. Thus for Day-one eggs, the between site variance rose with the distance the samples were apart, reaching a maximum at distances of 4 - 5 nm in an offshore direction and 10 nm in the alongshore direction. At larger distances, the variances declined forming a minimum again at 15-20 nm, which equates to these samples being on opposite sides of the patch (ie the diameter). These data suggest that to adequately sample pilchard eggs, sites should be no greater than these distances apart otherwise it is likely that patches of eggs will be missed completely.

The bootstrapping exercise demonstrated that obtaining more than the usual 3 to 4 samples (using the standard 5 x 15 nm grids) would be required in order to obtain a more precise mean value estimate for an area of 9 x 12 nm. The separation needs to be reduced such that samples are only 4 nm apart in an offshore direction and approximately 10 nm in the long shore direction. This would increase the numbers of samples in this size area to greater than 6 and therefore reduce the standard deviation and the confidence limits by approximately 50%. The increases in time required to increase the sampling frequency beyond a 4 x 10 nm grid would be prohibitive and less cost effective. However, further reductions in variance may be achieved if extra tows could be completed on an opportunistic basis during surveys if large numbers of eggs are observed by increasing sampling intensity in these areas.

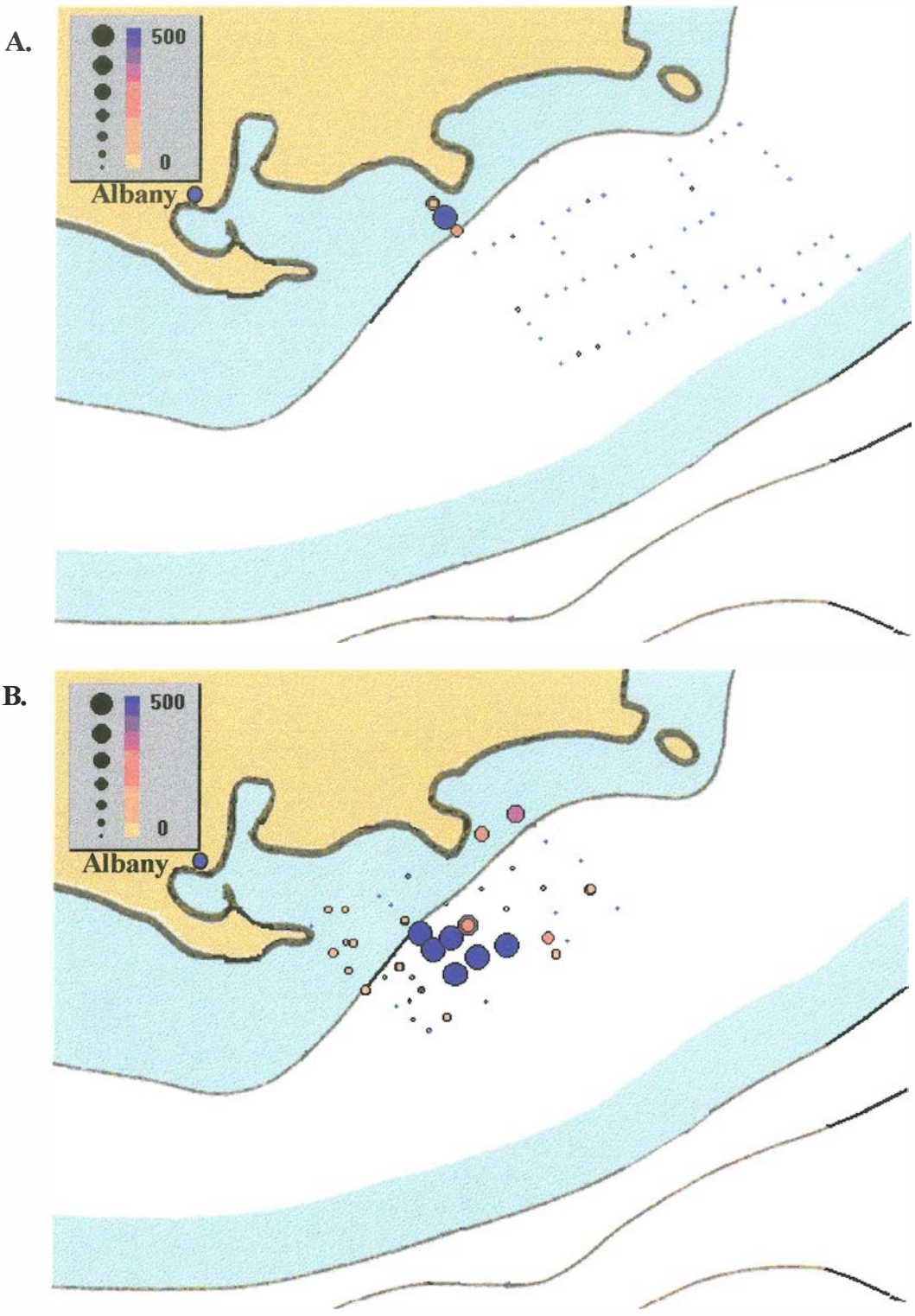


Fig. 6.1: Concentration of Day-one eggs collected on the two intensive surveys completed in (a) July 1993 and (b) January 1994

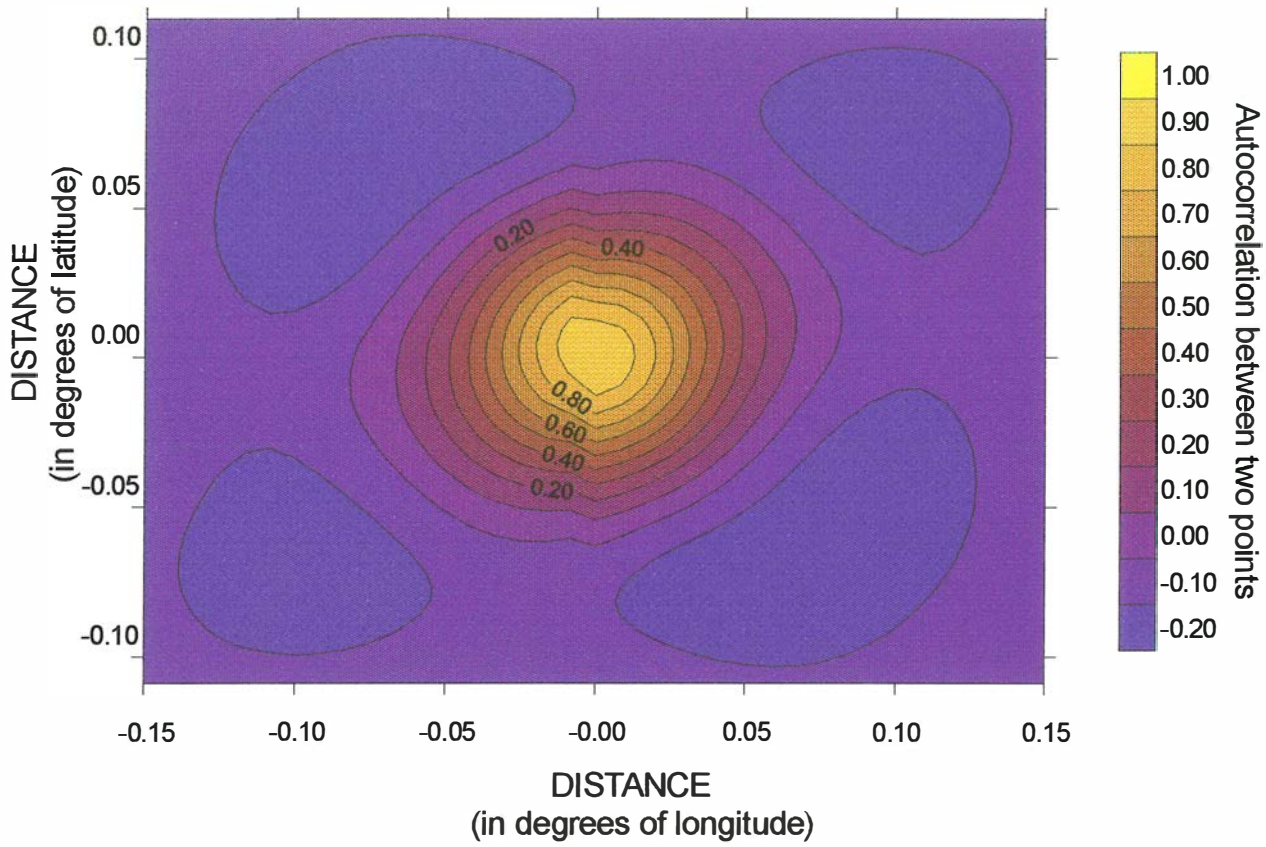


Figure 6.2: Correlogram for Day-one eggs collected in the January 1994 survey.

Non-Technical Summary

The WA pilchard fishery has expanded rapidly during the past 15 years. During this period there has been increased effort in the established areas of Fremantle and Albany with the new fishing areas of Bremer Bay and Esperance being further developed. The annual catch has increased to levels approaching 10000 t, and detailed research advice is now required for the management of these resources.

Gaining information on the stock size and structure of pelagic fish, such as pilchards, for use in their management is difficult. Their schooling behaviour makes the examination of commercial catch and effort data unreliable and the capture of adults for research purposes difficult. In addition, the restricted location of fishing operations in comparison to their extensive distribution often requires alternative research. Consequently, many stocks of pelagic fish are now monitored using the fishery independent techniques of egg/larval abundance and distribution indices.

The objectives of the present study were to document the distribution of the eggs and larvae of pilchards along the south and south-west coasts of Western Australia over a number of seasons and years to determine: (1) the structure of the stocks among the different areas where pilchards are caught, (2) provide estimates of the spawning biomass using egg production techniques, (3) examine the possible influence of oceanographic conditions on the location of spawning and the subsequent fate of the eggs and larvae.

A total of 8 plankton surveys were completed between July 1992 and July 1995 covering various portions of the coast from Adelaide to Fremantle with over 1000 plankton samples collected. From these data it was found that pilchards have a widespread, but patchy, distribution along the continental shelf waters of WA. The west coast and south coast stocks appear to be largely separate with a consistent gap in spawning between Cape Naturaliste and Torbay. The south coast possibly has a number of less isolated stocks. Separate spawning concentrations were consistently found off Albany and Bremer Bay, with Esperance having a peak spawning period a few months earlier than these two locations. Sampling across to Adelaide indicated that the WA spawning stock finishes at the western end of the Great Australian Bight (GAB). However, the substantial transport of eggs and larvae to the east when the Leeuwin Current is flowing, may result in eggs spawned near Esperance being advected across to SA by late larval stages. Similarly, those spawned near Albany often appeared to be advected over 100 km by the time they were early larvae.

Integrating the data collected on the area and intensity of spawning with adult reproductive characteristics allowed the calculation of the spawning biomass of pilchards. A number of separate estimates of biomass were calculated for each of the main fishing areas but the reliability varied greatly; those from Albany were the best, while those from the west coast the poorest. Biomass estimates for the Albany region ranged from 16000 - 32000 tonnes with a slight correlation between these values and the relevant computer model based estimates during the period. Substantial stocks of pilchards also occur off Bremer Bay and Esperance, with preliminary estimates

between 15000 - 30000 t range at each location. The data obtained on the west coast, however, were too imprecise to use.

Additional studies were completed to assist with the design and analysis of future surveys. Thus, the relationship between the rate of egg development and ambient seawater temperature was determined. The time taken to complete development varied between 32 - 50 hours over the temperature range of 16 - 21 ^{o}C. An examination of the vertical distribution of eggs and larvae showed that majority of pilchard eggs were spawned at about 30-50 m depth and then subsequently float to the surface. Finally, the dimensions of the patches of pilchard eggs was estimated using repeated sampling at close interval. This has allowed the determination of the optimal sampling distance which will be used in future surveys.

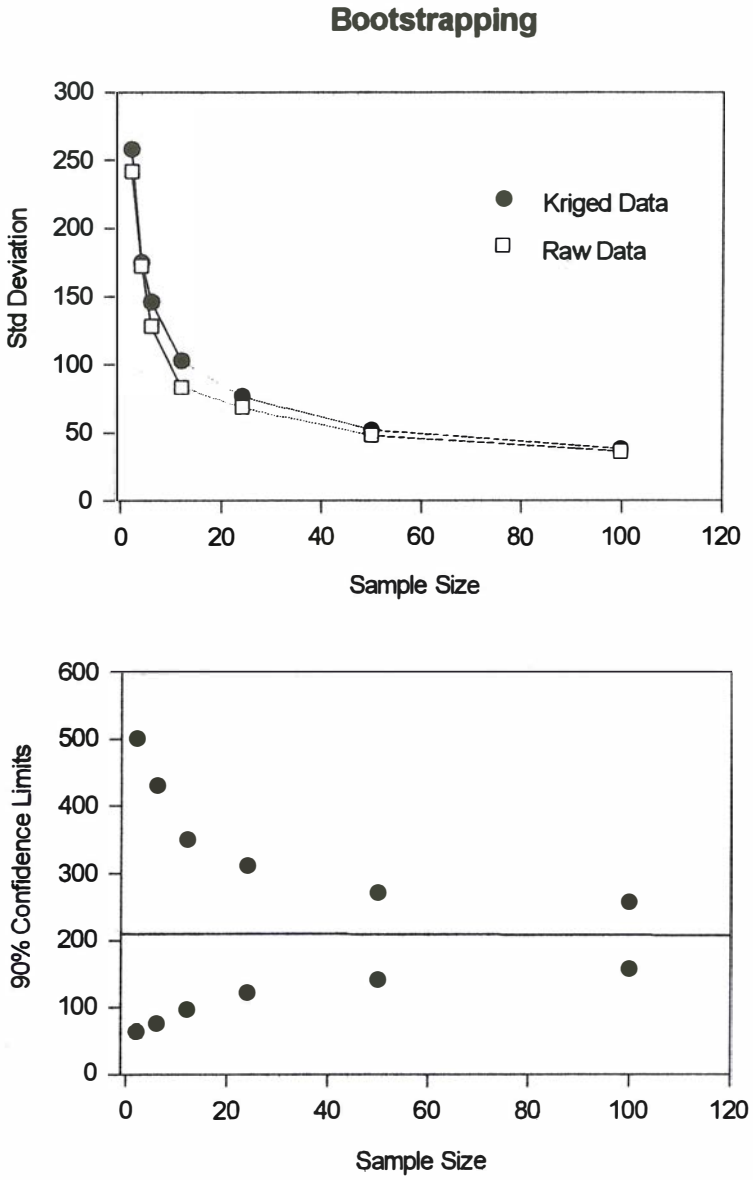


Figure 6.6: The standard deviation and 90% confidence limits for estimates of abundance calculated using 1000 simulations of between 2 and 100 samples of the kriged and raw data from the January 1994 survey.

7. Section 7: The Vertical Distribution of Pilchard Eggs and Larvae

7.1 Introduction

Understanding the patterns of vertical distribution for eggs and larvae is necessary for a complete description of the spawning and early life history of pelagic fish species. The vertical position of eggs and larvae will have an impact on their fate in a number of ways. First the rates of advection may be affected due to differential rates of current flow that occur with depth or the relative influence of winds closer to the surface. Vertical position may also impact upon the rates of mortality and growth due to the coincidence or avoidance of predators and food.

For the accurate interpretation of plankton samples obtained from vertical or oblique tows it is also essential to know to what depths to sample and where the majority of the eggs are located. The position in the water column may also influence the relative efficiency of sampling using plankton nets. Fletcher *et al.* (1994) found a substantial difference in the relative densities of different aged pilchard eggs between these two towing methods. Oblique hauls tend to collect relatively more Day-two eggs (those > 24 hr) and relatively less Day-one eggs compared to vertical tows. It was suggested that this difference could have occurred due to variations in the depth of pilchard eggs with age and the different towing profiles of the two methods.

There have only been a few studies on the depth distribution of pilchard eggs which were completed in North America and Europe (eg Ahlstrom, 1959; Coombs *et al.*, 1985); none have been done on Australian stocks. The oceanographic areas where pilchard eggs are found in Western Australia are very different from America, and

possibly even Europe, as the majority of eggs restricted to the continental shelf (< 100 m, Fletcher *et al.*, 1994) and there is almost no vertical water stratification (Cresswell, 1991 and see later).

The aims of this section were to obtain plankton samples at discrete depths to determine the vertical distribution of pilchard egg and larval stages. This work was conducted on the *RV Franklin* in a cruise from Adelaide to Albany during July 1994 using an EZ net (see Section 2).

7.2 Methods

The EZ net is a facility of the oceanographic research vessel *RV Franklin.* Up to 10 nets can be used on any one trawl and these are opened and closed independently using an on-board computer. Each net has a mouth opening of $1m^2$ with the mesh size used in this series of tows being 303 μm.. The speed of the boat, depth, water temperature, salinity and volume of water filtered are measured and displayed on a screen continuously during deployment. The design of the EZ net makes it unstable at depths shallower than 10 m, thus surface samples were obtained independently using additional nets towed from the side of the vessel. Both standard Bongo nets (0.6m diameter) with 300 μm mesh and a $1m^2$ surface net with 1000 μm mesh were deployed to sample this layer.

Sets of samples were obtained at a number of sites over the shelf and shelf slope between Port Lincoln and Albany. Between three and five nets were used for each EZ tow in addition to the surface net, this being dependent upon the depth at each location. The EZ nets were opened for 2 minutes whilst the surface net was towed, at the same speed as the EZ net for 10 minutes.

The amount of water filtered by each of the EZ nets was determined from the mouth area of the net, vessel speed and the duration of the tow (2 minutes). The onboard calculations appeared to be inaccurate giving calculated volumes which were less than one full net of water. The volume for the surface net was determined similarly and by using the flowmeter readings. Given the smaller size of the net 0.28 m^2 (compared to 1m^2), the increased duration (10 minutes) resulted in similar absolute amounts being filtered. These were resolved by adjusting the surface nets to a filtered volume of 200 m^3 which approximates the volume that would have been filtered by each of the EZ nets (ie. the boat travelled 100m in two minutes - 0.8m/s or 1.5 kt).

Eggs and larvae were sorted and staged as mentioned above (Section 2). Plankton volume was measured as the settled volume within a 2.5 cm diameter measuring cylinder.

Analyses for each of the 12 egg and 3 larval stages were completed using both absolute and proportional values located at each depth. Proportional values were calculated as the number present in each depth category in comparison to the total number collected in all samples from the site. Similarly the depths used were either absolute depths or expressed as a percentage of the bottom depth. These combinations allowed comparison among different depths and also among areas with different abundances of the pilchard planktonic stages. In some analyses, pilchard eggs were combined into four categories (stages 1-4; 5-7 8-10 and 11-12) or into Day-1 and Day-2 categories.

7.3 Results

7.3.1 General

A total of 33 EZ net tows were completed which provided 147 net samples for analysis. The bottom depths sampled varied from 49 - 130 m with net samples taken to 100 m where possible.

The volume of plankton collected in each tow is shown in Fig 7.1a. The largest amounts were seen at the surface with up to 33 cm of plankton collected. Pilchard eggs were collected at most depths down to 75m with none found below this depth. Pilchard larvae were also found to 70m but there were less at intermediate depths. Other fish larvae were found from the surface down to 100 m.

7.3.2 Pilchard Eggs

Analyses were only completed for tows in which at least one pilchard egg was collected to avoid biasing the results with numerous zero values. Thus only 23 of the 33 stations were used.

Stages: The numbers of eggs of each developmental stage collected are shown in Fig 7.2. Stage 3 eggs were most common at 30 - 60 m depth, stage 4 and 5 eggs were common at all depths from 0 - 60 m. Most older stages were, however, generally restricted to depths shallower than 40 m with stage 11 eggs almost all shallower than 20 m. The depth zone where stages were most abundant was the surface for all but stages 3 and 5.

Half Day Intervals: There were slightly different patterns between the actual density and relative density for each of the four categories of pilchard eggs with relative depth (Fig. 7.3a). There were also some differences in their position between daytime and

nightime tows. Thus, for stages 1-4 (0 to 12 hours old), daytime tows collected the majority of eggs at 30% of the depth with another minor peak at 60%. At nightime, the majority of eggs were collected at a depth that was 60% of the bottom depth. For stages 5-7 most eggs were caught between 15 m and the surface in both day and night tows with a minor peak at the 50 - 60 % depth. Most eggs in the Day-two categories (ie. Stages 8-10 and 11-12) were collected at the surface except for a peak for stages 8-10 at 60% of bottom depth during the day and 30% depth during nightime tows.

Day Intervals: The vertical distribution of Day-one and Day-two eggs are shown in Fig. 7.4. Using actual depths, Day-one eggs were most abundant at the surface, with a secondary region of large abundance between 35 and 60 m depth, declining to zero abundance below 80 m (Fig. 7.4a). Using relative depth, the largest average density was at 60% of the total depth. This depth had the largest mean value even when one large catch of 1500 eggs was removed, (see arrow Fig. 7.4b).

The vertical distribution of Day-two eggs using both actual and relative depth showed similar patterns. Thus most were located at the surface with a steady decline in abundance with increasing depth but a small increase at the 60% depth (Fig. 7.4a,b).

7.3.3 Pilchard Larvae

Yolk-sac Larvae: During the daytime, most of this stage were collected at the surface with mean capture rates of 90 per tow with relatively few collected at deeper locations (Fig. 7.5). During night tows, the maximum numbers of these larvae were collected at 30 m depth but the mean capture rate was only 18 per tow.

Early Post Larvae: The mean rate of capture for these larvae during the day at the surface was 166 per tow declining to zero at 80 m. At night, the maximum capture rate was at a depth of 15 m with a mean of 75 per tow.

Late Post Larvae: The largest numbers of this stage were collected at the surface during the day and at night from 30 m depth. Whilst the actual rate of capture during the day was greater than at night, late post larvae made a higher proportion of the catch at night (15%) compared to the day (6.5%).

7.4 Discussion

7.4.1 Pilchard Eggs

Pilchard eggs and larvae off the south coast of Australia were not uniformly distributed throughout the water column. Furthermore there were substantial differences in the relative position among the various stages of development.

We found no pilchard eggs below 80 m depth. Therefore, plankton sampling probably only needs to be completed to 70 m depth to adequately sample pilchard eggs. A relatively shallow distribution has been found elsewhere for *Sardinops sagax* with both Ahlstrom (1959) and Konishi (1980) finding either no or few eggs below 70 m off California and Japan respectively. Similarly, sampling for the eggs of the northern anchovy, *Engraulis mordax* is only completed to a depth of 70 m (Lasker, 1985).

The youngest stages of pilchard eggs found in this study (Stage 3) were found most commonly at approximately 40 - 50 m depth (or 60 % of the bottom depth). This indicates that pilchard spawning off southern Australia probably occurs at or near these depths, not near the surface. This is consistent with the prediction from Fletcher

et al. (1994) who found larger catches of Day-one eggs in vertical tows compared with oblique tows. The latter method tends to under-sample deeper layers of the water column due to the path of the net.

Ahlstrom (1959) also found that early stage *S. sagax* eggs were located in deeper layers at 28 - 68 m during the day and between 17 - 55 m at night. Similarly, Cushing (1957) found most stage-I *Sardina* eggs (which equates to our early Day-one eggs) were found between 40 and 60 m. Strangely, from these data it was concluded that pilchards must spawn at midday and the eggs sank, but it is more likely that they had only recently been spawned. Coombs *et al.* (1985) found when there was no thermocline in the water column, the distribution of *Sardina* eggs was bimodal with many eggs found at the surface and upper layers and another mode from 20-54 m. Unfortunately the eggs were not staged so it cannot be ascertained if the deeper mode comprised early stages. Konishi (1980), however, found most early stage eggs of *S. sagax* off Japan were located near the surface.

In the present study, older egg stages were mostly located in surface layers. Eggs of the closely related *Sardina* tend to be positively buoyant (Coombs *et al.*, 1985) thus pilchard eggs probably float to the surface after spawning. Thus it is common for large numbers of older eggs to be found in this layer (Ahlstrom, 1959, Cushing 1957, Konishi, 1980) but with some tendency for the final stages to sink again (eg Konishi, 1980). The concentrations that are seen in the surface layer make it imperative that when sampling, the net is not dragged horizontally at the surface at the end of a vertical tow because this could grossly inflate the rate of capture.

7.4.2 Pilchard Larvae

During the day nearly all pilchard larvae were most abundant in the surface layers. At night there was a tendency for many of the pilchard larvae to be deeper at 15 - 30 m. There was no accompanying alteration in the position of other components of the plankton.

The catch rates of pilchard larvae were greater during the day than at night for all stages. This is in contrast to all other studies where night time capture rates of larvae have been greater for both pilchards (eg Ahlstrom, 1959) and other species (see Heath, 1993 for review). No explanation is available for this pattern.

7.4.3 Conclusions

This study has confirmed the need to ensure that the path of the net is as vertical as possible for quantitatively sampling pilchard eggs. The difference in the vertical position of the egg stages means that under-sampling of early stages will occur if the deeper regions are not uniformly sampled. Furthermore, the depth that the net samples to may also impact upon the total numbers of eggs collected

These possible impacts have implications for some of the egg production estimates obtained given that in some surveys the path of the nets was not able to be kept vertical, particularly during windy conditions. The recent change to the smaller and relatively heavier CalVET net appears to have ameliorated this impact.

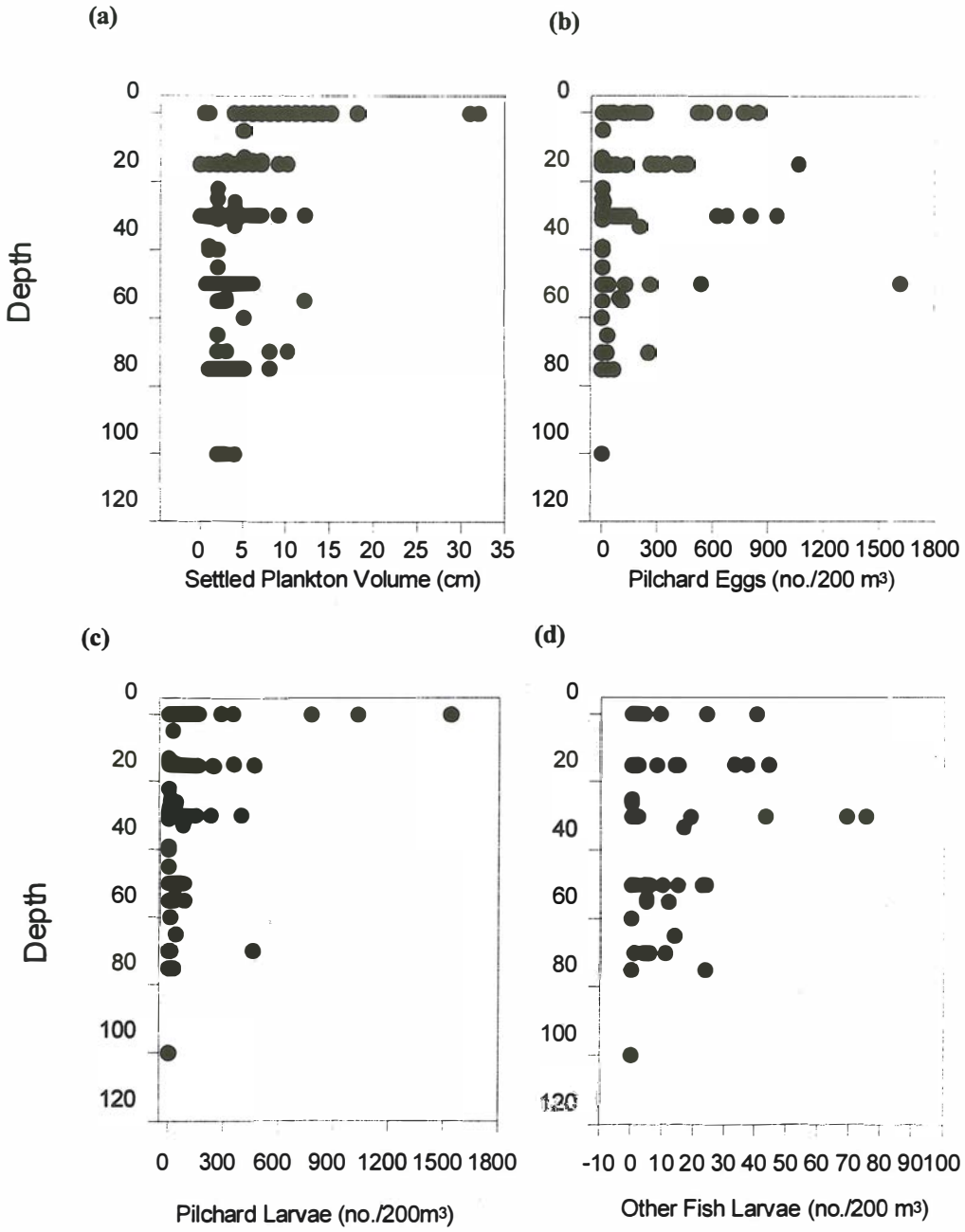


Figure 7.1: The vertical distribution of (a) settled plankton volume (cm); (b) all pilchard eggs, (c) pilchard larvae, (d) other fish larvae

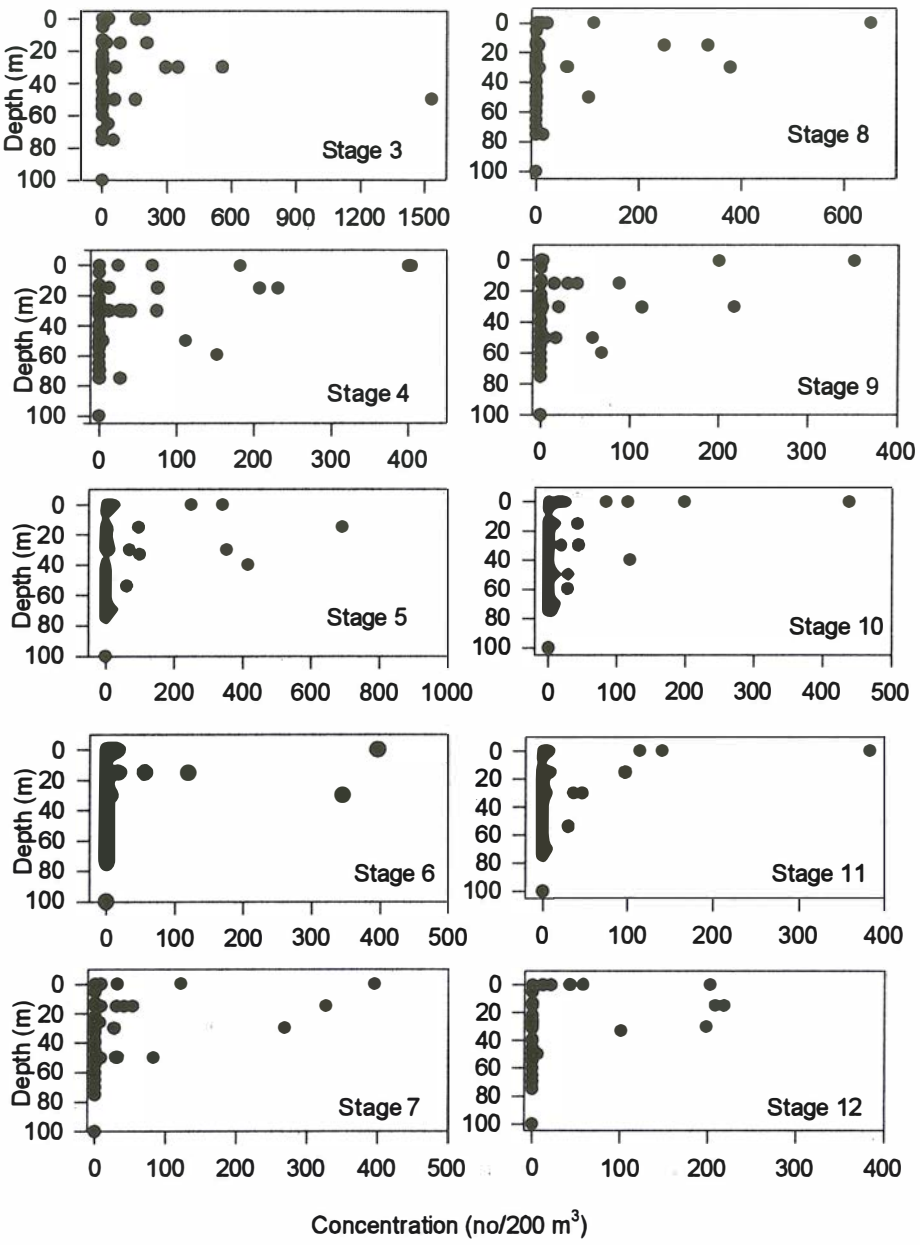


Figure 7.2: The vertical distribution of each of the pilchard egg stages against absolute depth collected by the EZ net.

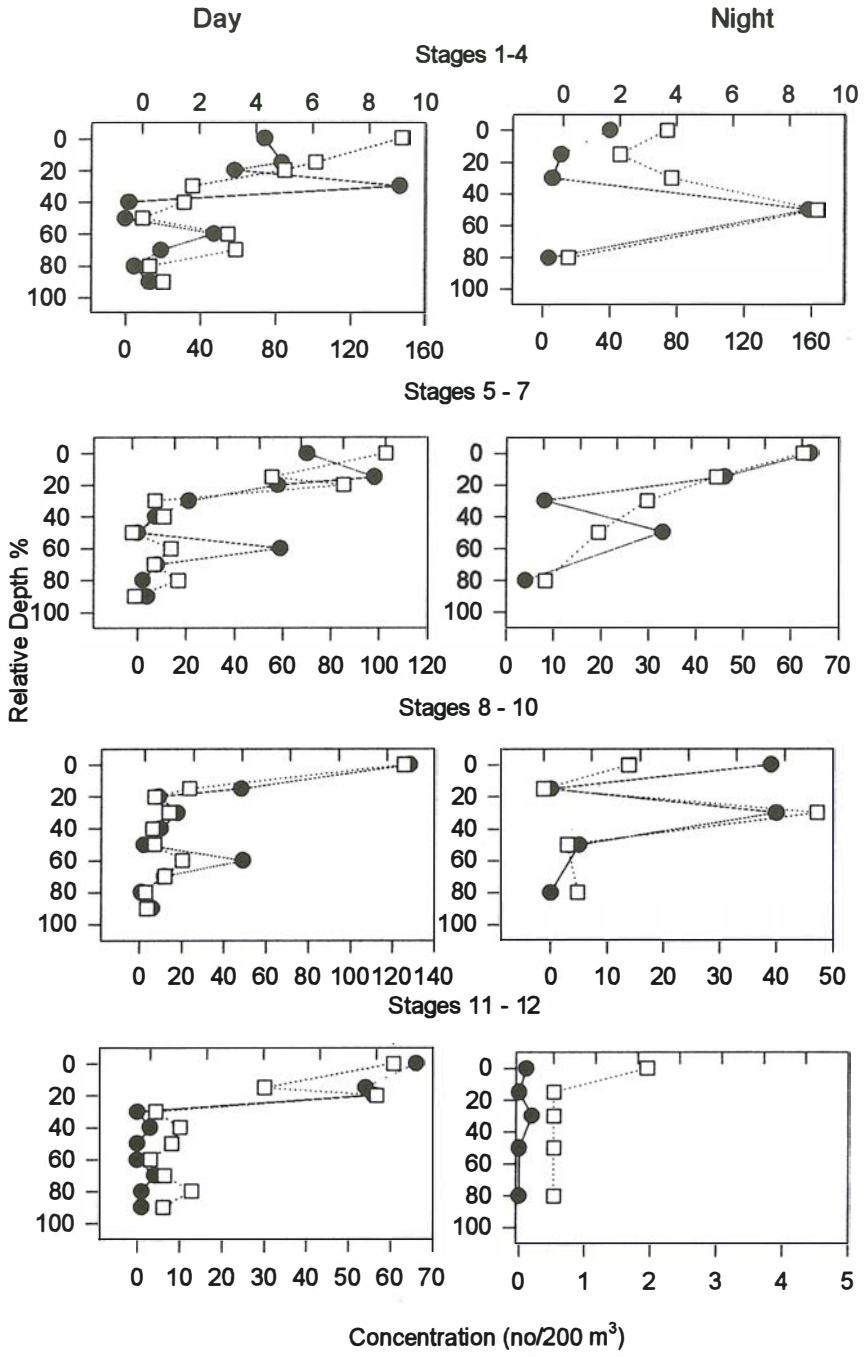


Figure 7.3: The vertical distribution of the 4 half day stages of pilchard eggs using absolute (solid line) and relative (dashed line) concentrations plotted against relative depth.

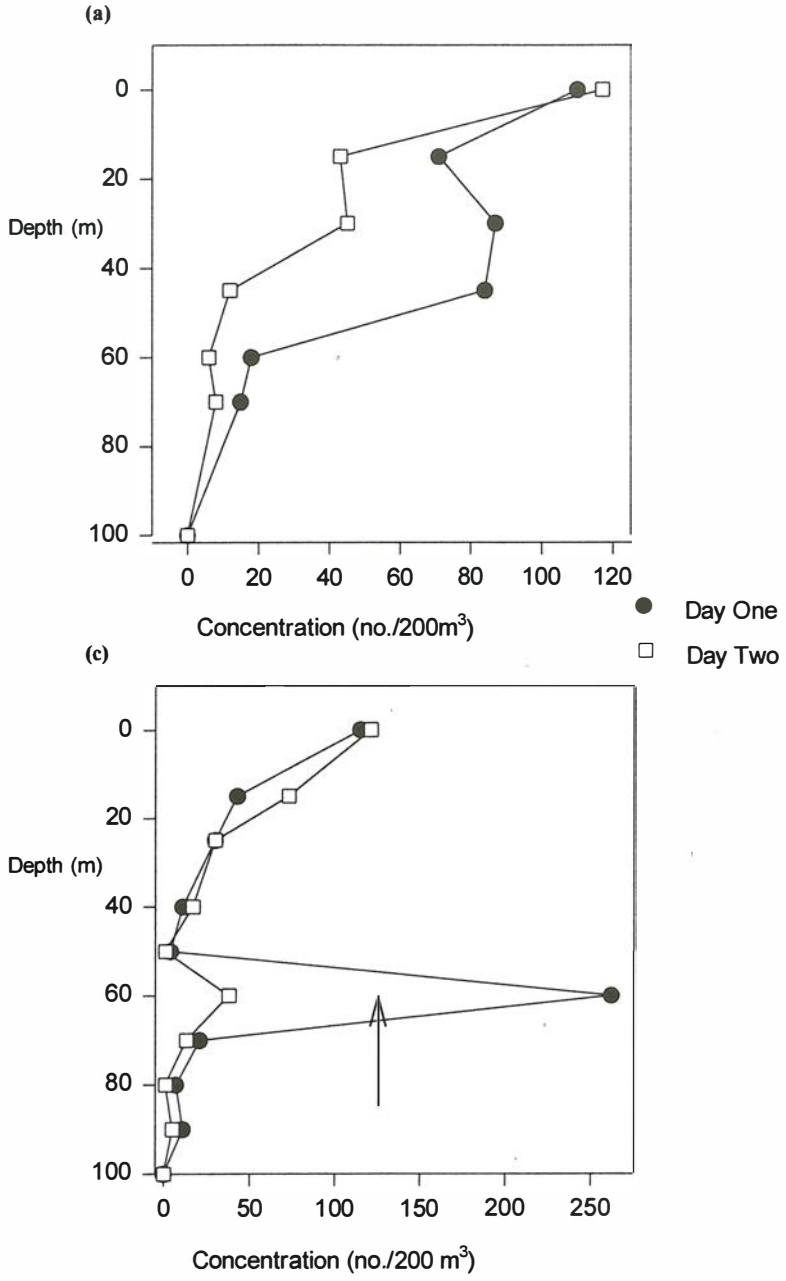


Figure 7.4: The vertical distribution of Day-one and Day-two pilchard eggs plotted against (a) absolute and (b) relative depths. The arrow indicates the adjusted mean after the exclusion of one sample with an extraordinarily large value.

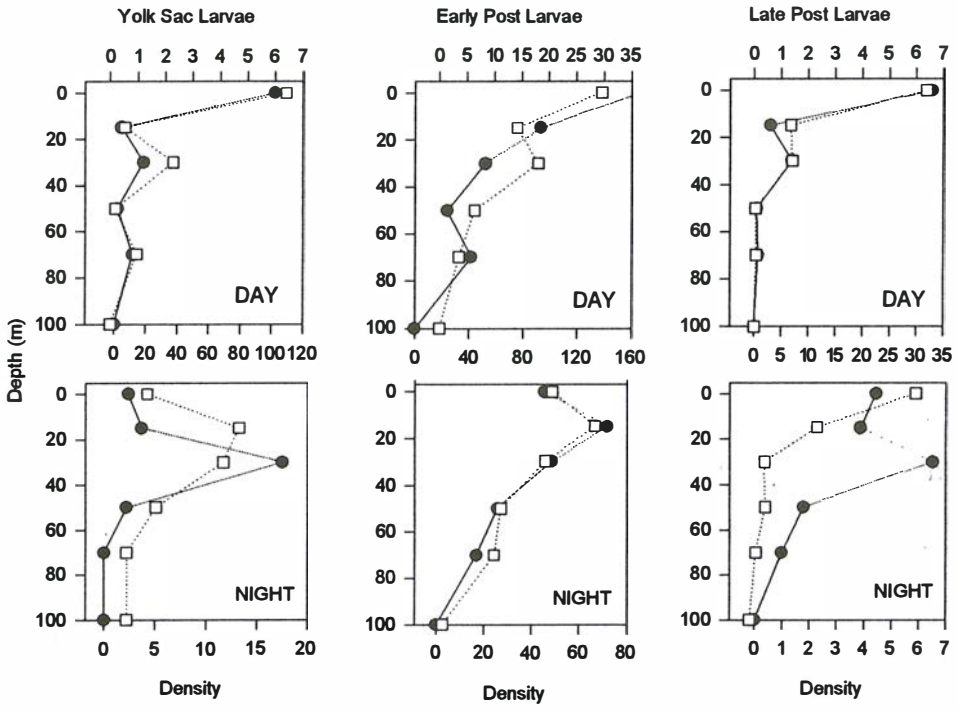


Figure 7.5: The absolute (solid line) and relative (dashed line) concentration of the 3 pilchard larval stages caught by the EZ net plotted against depth for both day and night tows.

8. Section 8 Comparison of Capture Probability and Efficiency of CalVET nets and Various Bongo Nets

8.1 Introduction

During the progress of this project a number of changes were made to the methods of sampling. These changes were instigated as more information was gathered on the sampling characteristics of nets and the density of pilchard eggs. The towing methods began using standard Bongo nets of 500 μm mesh towed in a double oblique method in which each tow lasted 10 minutes. This was subsequently changed to using standar Bongo nets of 500μm and 300μm mesh but towed in a vertical fashion (sampling predominantly on the way up) and finally to the use of CalVET nets (Smith *et al.*, 1985) of 300 μm mesh (only sample on the way up). Such variations require some investigation of what the impact each of these methods had on the number of positive tows that are generated and the relative density of the Day-one, Day-two eggs and larval stages.

It has already been shown (Fletcher *et al.*, 1994) that the method of towing standard Bongo nets (vertical or oblique) can affect the capture rates of the different stages of eggs. Such variation was likely to be associated with the different depth distribution of eggs of different stages (see section 7). It was also possible that the change in the use of nets from the standard Bongo to CalVET design may also have had some affect on the probability of capture and the relative capture rates of pilchard eggs. This knowledge is vital for the analysis and comparison of the results for biomass estimation and also for comparison among cruises that used different towing methods. Similarly, a different mesh size is known to affect the capture rate of eggs and larvae

of anchovies (Nancy Lo, pers comm) due to differential extrusion. Consequently some understanding of the effects of using 300μm or 500 μm mesh was required.

8.2 Methods

8.2.1 500μm vs 300 μm Mesh

In the two patch studies completed in the Albany region (Section 6), the standard Bongo nets used on each boat had different mesh size. One net had a mesh size of 500 μm, the other net had 300 μm mesh. The contents collected by each of these nets were kept separately for analysis. Furthermore, the data collected on the two boats used for this survey were analysed separately with the different numbers of each egg and larval category assessed using pair-wise t-tests and regression analyses calculated using SigmaPlot© Version 5. The total amount of plankton collected and the time taken to sort the pairs of samples were also investigated.

8.2.2 CalVET vs Standard Bongo

During the July 1995 survey in the Albany region, hauls of both the CalVET and standard Bongo nets were made at the same stations on 22 occasions covering sites from inshore to offshore locations. The samples were sorted independently with the numbers of all pilchard eggs, Day-one and Day-two eggs converted using the flowmeter readings and the relative mouth apertures into indices based on the filtration of 200 m^3.

Paired t tests and presence absence analyses were conducted using the "ANALYSIS" routines in MICROSOFT EXCEL© version 5.

8.3 Results

8.3.1 vs 500 *μm* mesh

Eggs: There was strong similarity in the numbers of eggs of both ages collected in the two adjacent bongo nets (Fig. 8.1). Almost identical counts of eggs were recorded in each net at both low and high abundances. Thus, the slopes of the regression lines were almost exactly 1.0 for three of the four comparisons with high correlation coefficients and with the paired t-tests indicating that there were no significant differences (Table 8.1). The only exception was for the Day-two eggs on transect 1 where the very tight relationship had a regression slope of 1.36 which resulted in a significantly greater number of this stage collected in the 300 μm mesh net.

Larvae: There were large differences in the relative numbers of most larval stages collected in the two different mesh nets with the 300 μm mesh nets catching far more larvae then the 500 μm mesh (Fig. 8.1). On transect 1, the slope of the regression was 5.65 with a tight fit about the line whereas on transect 2, the slope was higher at 8.26 but with more variability (Table 8.1). The paired t tests both confirmed the significant difference in catch rates (Table 8.1).

Early post-larvae were also collected more in 300 μm mesh but the differences were smaller with the slopes for the two transects similar at 2.74 and 2.23 respectively with a significant difference in catch rates (Table 8.1).

The capture efficiencies of late post larvae again varied between transects with transect 1 having significantly more caught in the 300 μm net but on transect 2 there was no significant difference between nets (Table 8.1).

8.3.2 Sorting Time

Records were kept of how long it took to sort the samples and a comparison between the mesh sizes was made. The 300 μm mesh nets collected four times more material than the 500 μm mesh nets (Table 8.2). Similarly the time taken to sort the 300 μm mesh samples was approximately 3 - 4 times longer.

8.3.3 CalVET vs Standard Bongo samples

Of the 22 sites where both tows were completed, pilchard eggs were only found at 14 (Table 8.3). Pilchard eggs were collected more often in the larger standard Bongo net samplers (0.52 m^2) than the smaller CalVET net (0.1m^2). Thus there were two sites where the standard Bongo nets collected pilchard eggs but none were found in the CalVET nets; the reverse was not observed.

The difference in presence/absence between samplers was largest for Day-one eggs. Only 7 of the 11 stations where the standard Bongo net caught Day-one eggs was the CalVET nets successful (Table 8.4). The stations where this stage was missing in the CalVET tows were, however, restricted to areas where their density was low with a mean index of 24 which is less than the minimum for the CalVET net (Table 8.4). When present, the numbers of Day-one eggs were similar in both nets, the slight trend to increased numbers in the CalVET which was not significant (Table 8.3; Fig. 8.2).

There was very little difference in either the rates of capture or the presence/absence of Day-two eggs between the two sampling methods (Table 8.3, Fig. 8.2). Nonetheless, the only station where Day-two eggs were present in the standard Bongo sample but missing from the CalVET net had a relatively low index (17.1; Table 8.4).

8.4 Discussion

8.4.1 500μm - 300 μm mesh

The close relationship between the numbers of eggs collected by the two mesh sizes on the one frame indicates that there is very little small scale (1 metre) variation in egg densities. The lack of a consistent difference in the capture rate also shows that there is little extrusion of pilchard eggs through the 500 μm mesh. Given that pilchard eggs are all greater than 1.1 mm this was to be expected.

However, the change in relative diameter of pilchard larvae after hatching makes this stage vulnerable to extrusion. The 300μm mesh nets captured between 5 - 8 times more yolk-sac larvae compared to the adjacent 500μm mesh net. The relationship appeared to vary with the speed of the tow, thus no simple calibration factor exists. The rate did decline for older stages and was almost insignificant for late post larval stages. Consequently, whilst a general understanding of the larval distribution of pilchards can be obtained using 500 μm mesh nets, for more precise estimates of abundance, 300 μm mesh is required. The greatly increased time required for sorting samples using this smaller mesh size, should however, be incorporated into any survey planning.

8.4.2 CalVET vs Bongo

There was a reasonably close relationship between the catch of pilchard eggs by the two types of nets. The regressions were not, however, as close as found previously for surveys using two standard bongo net samples at the same site (see above). Furthermore, there was a difference in the presence/absence data, particularly for Day-

one eggs, with the larger standard Bongo net collecting this stage at 11 sites and the CalVET net only at 7 sites.

The large difference in the size of the nets, and consequently the amount of water filtered, means that the probability of capture will be substantially lower for the CalVET net. Thus there will be a number of sites that the bongos collect eggs where the CalVET does not. This was predicted by Smith & Hewitt (1985).

Additional sampling needs to be done to classify more precisely the percentage of CalVET tows that will be zero when pilchard eggs are present in the area. It also suggests that it may be useful to use standard Bongo nets in areas where the egg density is thought to be low to increase the chances of obtaining a more accurate spawning area estimate.

Pilchard Stage	Regression			Paired t-test		
TRANSECT 1	**Slope**	**Intercept**	R^2	*t*	*P*	**df**
Day-one eggs	1.1	-1.7	0.97	1.008	0.32	29
Day-two eggs	1.36	1.56	0.98	2.83	0.008	29
Yolk-Sac Larvae	5.65	5.8	0.87	4.35	0.0001	29
Early Post-Larvae	2.74	10.2	0.8	5.09	0.00001	29
Late Post-Larvae	1.3	3.1	.71	2.52	0.017	29
TRANSECT 2	**Slope**	**Intercept**	R^2	*t*	*P*	**df**
Day-one eggs	1.01	-2.8	.95	0.27	0.78	19
Day-two eggs	1.00	4.6	.97	0.49	0.62	19
Yolk-Sac Larvae	8.26	101	.27	4.34	0.0003	19
Early Post-Larvae	2.23	23	.58	4.44	0.0003	19
Late post-Larvae	0.76	12	.79	0.57	0.577	19

Table 8.1: Paired t tests comparisons for pilchard stages collected in 300 and 500 μm mesh nets towed adjacently in two transects completed in January 1994.

	500μ mesh		300μ mesh	
	Mean	Std	Mean	Std
Plankton Volume (cm)	2.83	1.8	10.28	3.16
Sort time (min)	23.7	11	80.21	32

Table 8.2: Comparison of plankton volumes collected and sorting time between 300 and 500 μm mesh nets.

	Bongo			CalVET			Paired T Test	
	Positive	Mean	STD	Positive	Mean	STD	t	P
All eggs	14	314	738	12	459	1340	-0.81	0.42
Day-one	11	171	567	7	325	1327	-0.903	0.37
Day-two	12	142	497	11	134	325	0.18	0.85
Log(Day-one)		0.91	1.07		0.70	1.31	1.47	0.15
Log(Day-two)		0.99	1.04		1.05	1.14	-0.54	0.58

Table 8.3: Comparison of egg indices for pilchard eggs in Bongo nets and CalVET nets collected at the same sites.

	Day-one Eggs		Day-two Eggs	
	CalVET Negative	CalVET Positive	CalVET Negative	CalVET Positive
Mean	24.1	524	17.1	150
Min	7.1	9.5	17.1	9.6
Max	53	2616		2316

Table 8.4: Comparison of egg indices for standard Bongo net samples at sites where the CalVET nets caught (positive) or did not catch (negative)pilchard eggs.

Note: When one egg is found in a CalVET net the index per 200 m^3 is approximately 30.

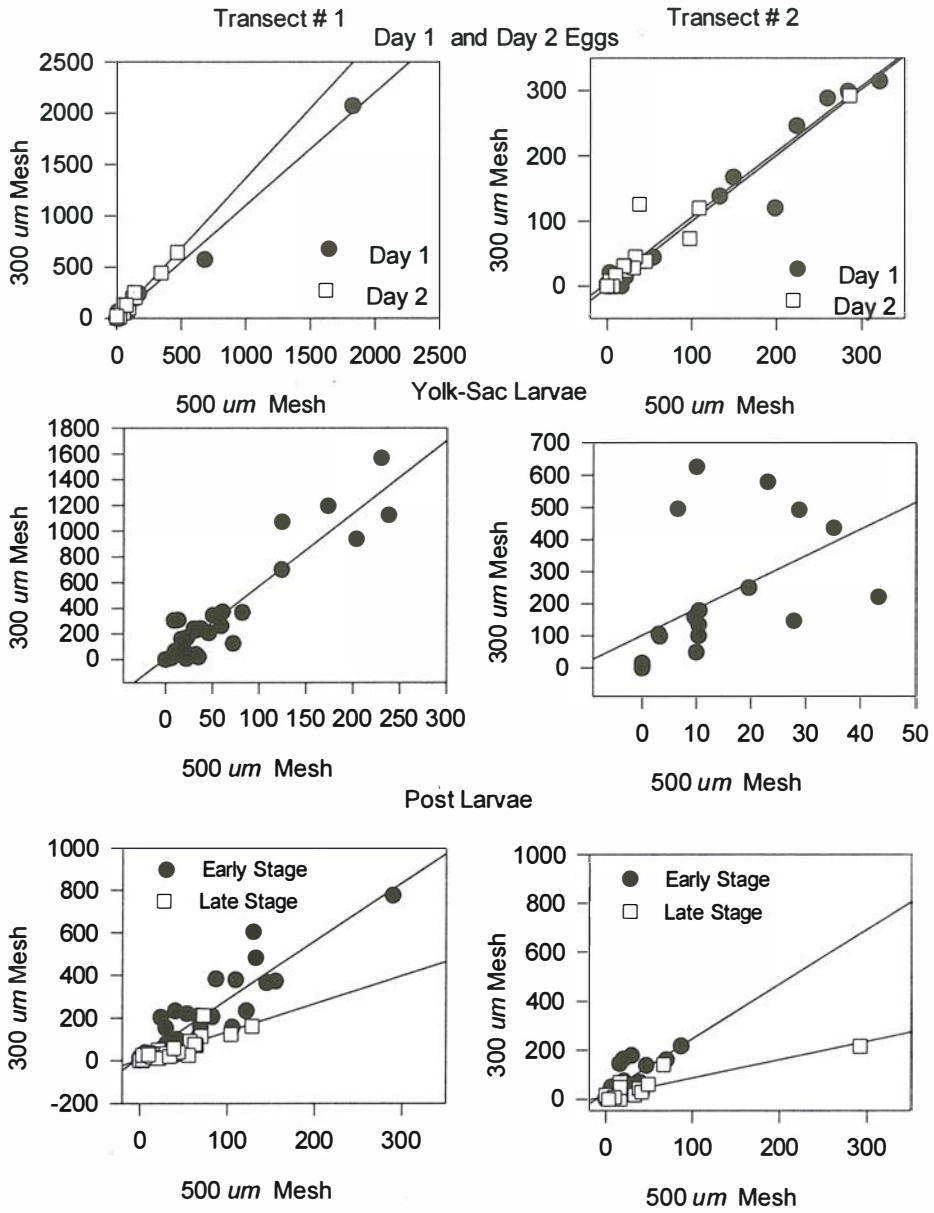


Figure 8.1: Comparison between the numbers of eggs and larvae captured in nets of two mesh sizes (300 and 500 micron) towed on the same frame for two separate surveys in the same area on the same day.

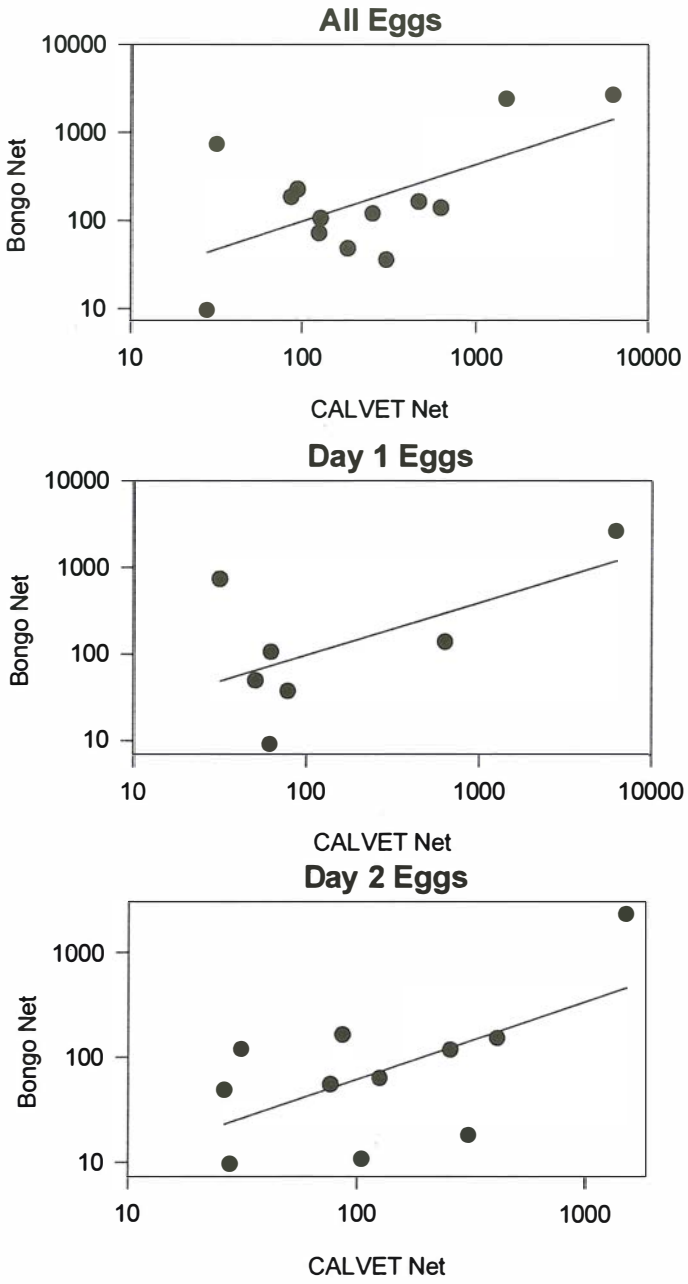


Figure 8.2: Comparison of the relative numbers of pilchard eggs per 200m^3 filtered water collected by standard Bongo nets and CalVET nets towed in the same locations.

9. Section 9 General Discussion (Benefits)

This study has highlighted the benefits of focused plankton surveys to assist with the investigation of commercially exploited species. It was not only successful in achieving all the original aims and objectives, but a number of additional topics were able to be investigated. The information gained covered technical improvements in sampling and survey methods, the patterns of distribution for pilchards eggs and larvae over a large area of Australia, and the calculation of pilchard biomass for a number of regions.

Our procedures for completing and analysing plankton samples have been greatly improved as a result of this study. Determining at what depth pilchards tend to spawn has helped us understand the variations in the relative catch rates of different aged eggs using different towing methods. Because the majority of early stage eggs are deeper in the water column and most older eggs are near the surface requires a strong effort to ensure that the trajectory of tows is as vertical as possible. The close observation sampling has demonstrated that there is a large degree of spatial cohesion in the distribution of pilchard eggs. The optimal distance between samples has therefore been determined and will be used in future surveys. Finally, the impact of the various of mesh sizes and nets used on the relative capture rates of eggs and larvae have shown that mesh size of 300μm or 500μm has no impact on eggs but the larger mesh allows the extrusion of up to 80% of early larval stages. The size of nets does not affect the capture rates of eggs when present, but the smaller CalVET nets have a

lower probability of capturing any eggs. The level of this effect requires further investigation. However, the benefits of using these small nets make this the preferred net for future use both in terms of their ease of use on board the vessel, the more quantitative way the sample (ie: only sampling in one direction) and the speed of sorting due to the low volumes of material collected.

The frequent and extensive plankton surveys which were completed during this study form one of the best datasets for the Australian coast. They have shown that pilchards have a widespread distribution in WA with there appearing to be a number of repeatable patterns in relative spawning intensity from which information on the separation among stocks could be inferred. Thus the pilchards on the west and south coast appear to be largely separate stocks with a consistently large gap in spawning between Cape Naturaliste on the lower west coast and Torbay, west of Albany. With this information we will be able to treat the fisheries in these two regions independently for management purposes.

On the south coast, the differences in the timing and concentration of spawning indicate that the adult stocks in this region may also to some extent be separated. Thus, Esperance has a different peak spawning time to Albany and Bremer Bay, and only Albany has both a summer and winter period of spawning. This supports the current management of these regions whereby separate quotas are used for each area based upon the assumption that the adult stocks are largely separate.

Sampling in the GAB and further east showed that this region does not appear to form part of the WA spawning stock. Nonetheless, the variable level of advection for the eggs and larvae during winter, dependent upon the strength of the Leeuwin Current suggests that there is probably substantial exchange between all south coast locations in some years. How this variability impacts upon subsequent recruitment back into the fishery will need to be monitored closely.

The calculation of biomass values using the data collected on egg abundance and distribution has been of great benefit to the management of the fisheries. The numerous estimates now available for the Albany region were all similar to the estimates calculated from the computer model. Thus, we are now more confident about our knowledge of the stock size in this area.

The stock size values calculated for Bremer Bay and Esperance, whilst not as precise as those for Albany, were sufficient to show that there are large stocks of pilchards in both zones. In Esperance, the knowledge from the plankton study has been instrumental in getting an increase in the TAC from 1200 to 1800 t. Further increases are possible.

Unfortunately, the information collected for the west coast was inadequate to allow any firm estimate of stock size for this region. What this highlights is the need to restrict surveys to a size that can be managed with the available staff. Thus in future, only one section of coast (ie. West coast, Albany/Bremer Bay; Esperance) will be surveyed at a time. This should allow the best chance of the obtaining sufficient

samples such that all parameters can be estimated, and with a reasonable level of precision.

The information related in this report has already been communicated to industry through verbal and written reports. The conclusions have been used to assist with development of management plans for the fisheries.

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11. Section 11 Appendices

11.1 Larval Otolith Analyses

11.1.1 Introduction

Daily growth rings in the sagittal otoliths of larvae have been established for a variety of clupeoids (Panella 1971, 1974; Brothers *et al.*, 1976; Struhasker and Uchuyama, 1976; Methot and Kramer, 1979; Thomas, 1985; Hayashi *et al.*, 1989). The deposition of the initial increment occurs a number of days after and may coincide with complete yolk-sac absorption and the onset of feeding hatching (Methot and Kramer, 1979; Townsend and Graham, 1981; Hayashi *et al.*, 1989). Daily growth rings have been used to determine the growth rates (Methot and Kramer, 1979; Lough *et al.*, 1982; Castillo *et al.*, 1985; Thomas, 1985; Anguilera *et al.*, 1986; Fukuhara and Takao, 1988; Palomera *et al.*, 1988; Thorrold, 1988; Hayashi, 1989; Alvarez and Morales-Nin, 1992; Jordan, 1994) and age structure (Townsend and Graham, 1981; Jones, 1985; Thorrold, 1988; Alvarez and Morales-Nin, 1992) of larval populations. However, the number of days to the onset of increment formation and growth rates may vary as a function of temperature and/or food type and availability (Zweifel and Lasker, 1976; Methot and Kramer, 1979; Jones 1985 and references therein).

11.1.2 Methods

Larvae were collected aboard the CSIRO research vessel the *R.V. Franklin* during the winter 1994 plankton survey from Adelaide (South Australia) to Albany (Western

Australia).The tows were carried out from 7-7-94 to 23-7-94 and the location and number of each station is shown in Fig. 11.1. The larvae were collected using a 1 metre square, 1000 micron mesh net towed along the surface for up to 10 minutes. The cod-end contents were then placed in a sorting tray and any visible pilchard larvae were extracted and placed in 70% de-natured ethanol. The remaining portion of the sample was placed in 5% buffered formalin and seawater and later placed in 3% buffered formalin and seawater.

The samples were then sorted in the laboratory under a dissecting microscope and all remaining pilchard larvae were removed and placed in 70% ethanol. Standard length (notochord length in pre-flexion larvae) of all pilchard larvae were then measured. Larvae were placed on a microscope slide and sagittal otoliths were removed using two dissecting needles. The slides were air-dried and the otoliths fixed in an acetate based setting resin. Initially (stations 31,32 and 35) a coverslip was placed on the slide over the otoliths. This resulted in the otoliths being crushed as the mounting fluid dried and consequently no measurements were obtained for these otoliths. [*However, it was obvious that the majority of these damaged otoliths were large and had a greater number of rings than otoliths further west*]. All otoliths processed after this were mounted with no coverslip.

Otoliths were observed under a compound microscope and observed using transmitted light. Alternating light and dark concentric rings were obvious on otoliths (Fig. 11.2) and were assumed to be daily growth rings (as discussed by Panella 1971, 1974; Brothers *et al.*, 1976; Struhasker and Uchuyama, 1976; Methot and Kramer, 1979;

Thomas, 1985; Hayashi *et al.*, 1989). The microscope was attached to a desktop computer via a colour camera and the longest radius in the posterior field of each otolith was measured and the rings enumerated. The distance from the focus of the otolith to each ring was also measured.

In several of these otoliths the most central rings were obscured. This was mainly a result of inadequacies in the mounting fluid and, in some cases, tissue remaining attached to the otolith. It is therefore strongly recommended that otoliths be viewed and analysed in immersion oil within a few hours of extraction from the larvae. Once all daily rings were measured and counted a frequency histogram of distances of daily rings from the focus was constructed (Fig. 11.3). There were obvious peaks and troughs in this data which were taken as an indication of increment formation. An estimation of the number of rings obscured in each otoliths was obtained from the number of peaks in the frequency histogram which occurred prior to the first observed daily ring.

11.1.3 Results

The larval length versus increment unit (age in days) data were analysed using the generalised version of the von Bertalanffy growth function, which has the form:

$$L_t = \omega / k \, (1 - \exp(-k \, (t - t_0)))$$

where, L_t = length at age t; L_∞ = maximum larval size obtained; $\omega = kL_\infty$; k = growth constant; and t_0 = x-axis intercept. The data was also analysed using the Laird-Gompertz function which is generally used to analyse otolith daily increment data (Lough *et al.*, 1982; Palomera *et al.*, 1988; Thorrold, 1988). This function has the form:

$$L_t = L_0 \exp(A_0 / \alpha (1 - e^{\alpha t}))$$

where, L_0 = length at t = 0; A_0 = specific growth rate at t = 0; α = rate of exponential decay. Both of these equations yielded good fits to the observed data with an r^2 of 0.973 and 0.975 respectively.

The estimated values for the parameters of both equations using the observed data are shown in Table 11.1. Standard errors for these were estimated using a Jacobian matrix. The estimated lengths and growth rates using the Laird-Gompertz equation are summarised in Table 11.2. The growth rate is seen to increase to a maximum of 0.619 mm/d in the initial 10 days and then decreases after this point. This may be as a result of the Laird-Gompertz equation under-estimating growth at ages less than 10 days (Thorrold 1988).

The growth rate for the von Bertalanffy equation constantly decreases from an initial maximum of 0.945 mm/d. These results compare favourably with observed growth rates of other Clupeoid species from previous studies (see Table 11.3)

Growth of pilchard larvae were also compared between areas (Fig. 11.4). An analysis of the residual sum of squares (Chen *et al.* 1992) was employed and results are presented in Table 11.4. The results show a difference in growth pattern in all of the selected areas at the $p < 0.05$ significance level. The estimated value and associated standard error for each of the Laird-Gompertz equation parameters are presented in Table 11.5 and Fig. 11.5 shows the differences in the form of the estimated growth function. The differences between areas mainly arise from differences in length and specific growth rate at t=0, and from the maximum larval length obtained. These differences may however be an artefact of the range of lengths used to estimate the parameters of the function (Palomera *et al.*, 1988), thereby underestimating length for younger larvae in the Bight and overestimating length for older larvae in the Esperance area

The exact time of deposition of the initial increment has not been determined for *Sardinops sagax.* However in other Clupeiformes it coincides with absorption of the yolk-sac (Methot and Kramer, 1979; Lough *et al.*, 1982; Hayashi *et al.*, 1989). It is not possible, therefore, to obtain exact ages with the available data, however relative ages can be compared by comparison of the number of otolith increments and it may be assumed that:

age = (# of otolith increments) + (2 to 4 days of yolk-sac absorption)

When comparing the number of increments between areas it is obvious that larvae in the Bight area are generally much older than in the Esperance and Albany areas (see

Table 11.6). In the Bight area 42.11% of larvae have over 30 otolith increments. Esperance and Albany, however, have just 1.56% and 5.26% of larvae in this category respectively. Conversely, 81.25% and 57.89% of Esperance and Albany larvae have less than 20 otolith increments, whereas the Bight region has only 10.53% of larvae in this category and no larvae with less than 12 increments. Hence larvae in the Bight area are most likely at least 2 to 2.5 weeks old. The lack of younger larvae suggests that either they originated from a spawning ground remote from their area of capture or that spawning in this area is more discrete than is found in areas near Albany and Esperance areas (*reference to previous papers from this section*). The predominance of younger larvae in the Esperance/Albany regions suggests that larvae are spawned in this area and are then possibly carried to other areas.

11.2 Analyses of Other Fish Larvae

11.2.1 Methods

Larvae were collected during the July 1993 and July 1994 field trips (see Table 2.1). The larvae were collected using a 1 metre square, 1000 micron mesh net towed along the surface for up to 10 minutes (surface 1000 tows - July 1994 field trip only). In addition, a 600 mm diameter standard Bongo net with 500 micron mesh was used to conduct vertical hauls from not more than 70 metres or 3 m from the bottom (vertical 500 tows). The cod-end contents were placed in 5% buffered formalin and seawater and later placed in 3% buffered formalin and seawater. The samples were then sorted in the laboratory under a dissecting microscope and all fish larvae were removed and placed in 70% ethanol. All larvae were separated out into taxonomic groups. Some

larvae were then identified to the lowest taxonomic level possible. A list of some of the identified species are shown in Table 11.7.

There were notable differences between the larvae of the surface 1000 as compared to the vertical 500 tows. Larvae in surface 1000 tows were notably larger (and therefore more easily identified), and there were differences in the presence of major groups of larvae most abundant each sample set (Table 11.8). Surface 1000 tows are characterised by leptocephalus and morwong larvae as well as *Scombersox saurus* and *Gonorhyncus greyii*. Vertical 500 tows are characterised by Macrourids, Morids and Labrids as well as *Etrumeus teres*, *Scomber australasicus* and *Hyporamphus sp.*. The notable differences seen between the 1993 and 1994 vertical tows was the lack of Callionymid larvae in the 1994 samples.

11.2.1.1 Distribution

The distribution of some of the identified species are shown in Fig. 11.6, 11.7 and 11.8. Also notable from the data is the distinct dominance of deepwater species over near-shore species between Cape Leeuwin and Albany (Fig. 11.9). This distribution pattern is also reflected in the distributional abundances of pilchard larvae.

11.3 References

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Laird-Gompertz parameter	Estimated Value	standard error
L_0	2.995	0.031511
A_0	0.156	0.002276
α	0.80	0.001135
von Bertalannfy parameter	Estimated Value	standard error
ω	1.019	0.010987
k	0.043	0.001047
t_0	-1.246	0.071982

Table 11.1: Estimated values and standard errors of parameters for the Laird-Gompertz and von Bertalanffy Growth equations.

Age (increment number)	Estimated Length	Growth rate	
		mm/d	%/d SL
0	2.995		
1	3.478	0.483	16.13891
2	3.994	0.515	14.81517
3	4.537	0.543	13.60624
4	5.104	0.567	12.50126
5	5.691	0.587	11.49048
6	6.292	0.601	10.56521
7	6.904	0.611	9.71765
8	7.521	0.617	8.940803
9	8.140	0.619	8.228362
10	8.756	0.617	7.574646
15	11.694	0.559	5.023465
20	14.200	0.460	3.344928
30	17.656	0.260	1.49368
50	20.358	0.061	0.20230

Table 11.2: Estimated length and growth rate of larvae using fitted Laird-Gompertz function.

Author	Species	Growth Rate	Criteria	L_0	A_0	α
Present Study	*Sardinops sagax*	(see table 11.1)	15.4°C to 18.8°C	2.995	0.156	0.80
Methot and Kramer (1979)	*Engraulis mordax* (Northern Anchovy)	0.37 mm/d	15°C			
Townsend and Graham (1981)	*Clupea harengus harengus*	2.1 to 1.5 mm/week				
Lough *et al.* (1982)	*Clupea harengus* (Atlantic Herring)	0.25 mm/d 0.30 mm/d	10°C, at hatch 10°C, at 20 days	L_7 = 12.7	0.0267	0.03
Thomas (1985)	*Sardinops sagax* (Pilchard)	0.77 to 0.60 mm/d				
Thomas (1985)	*Engraulis capenensis* {=*E. japonicus*} (Anchovy)	0.44 to 0.77 mm/d				
Thomas (1985)	*Etrumeus whiteheadii* Round herring	0.53 mm/d				
Castillo *et al* (1985)	*Sardinops sagax*	0.4 mm/d 0.8mm/d	13mm 18.6°C, 13mm			
Aguilera *et al* (1986)	*Clupea bentincki*	0.37 mm/d 0.24 mm/d	12.5°C, 13 mm 26 to 37 mm			
Palomera *et al* (1988)	*Engraulis encrasiocolus* (West Mediterranian anchovy)	0.9 mm/d	20°C, 8mm	3.7973	0.1347	0.0590
Thorrold (1988)	*Herklotsichthys castelnaui*	0.57 mm/d 0.45 mm/d	4 to 5 days 14 to 15 days	5.159	0.104	0.075
Fukuhara and Takao (1988)	*Engraulis japonica*	0.41 to 0.62 mm/d	first 55 days			
Hayashi *et al* (1989)	*Sardinops melanostica* (Japanese sardine)	0.67 mm/d 0.58 mm/d	first 15 days 15 to 30 days			

Table 11.3: Comparison of growth rates and Laird-Gompertz Growth equation estimated parameters for several Clupeoid species.

Area	F-value	p-value
All areas	3.886	0.001
Bight and Esperance	2.798	0.043
Bight and Albany	3.164	0.027
Esperance and Albany	5.604	0.001

Table11. 4: Results of analysis of residual sum of squares (Chen *et al.*, 1988) to compare the estimated growth patterns of pilchard larvae between areas off the south coast.

Area	L_0	A_0	α
Bight	0.922	0.313	0.103
Esperance	4.350	0.114	0.078
Albany	2.382	0.201	0.091

Table 11.5: Comparison of Laird-Gompertz growth function parameter estimation for specific areas of the south coast.

	All	Bight	Esperance	Albany
Mean	20.65833	30.17105263	14.171875	15.8245614
Standard Error	0.748266	1.043843801	0.773620985	1.296835073
Median	20	29	14.5	17
Mode	15	24	10	4
Minimum	2	12	3	2
Maximum	53	48	31	38
Count	240	76	64	57

Table 11.6: Statistics for number of increments for all larvae collected and for specified areas along the south coast.

Taxonomic Group	Tow Type and Date		
	surface, 1000µm July, 1994	vertical, 500µm July, 1994	vertical 500µm July, 1993
Thamnacomes sp. Leatherjacket	*		
Scombinycthes granulatus Rough leatherjacket	*		
Scombersox saurus Saury	*		
Gonorhyncus greyii Beaked salmon	*		
Cheilodactylus nigripes Magpie perch	*		
Scorpis sp Sweep	*		
Cheilodactylus sp. Morwong	*		
Nematodactylus macopterus Jackass Fish	*		
Cheilodactylus rubrolabatius Red-lipped morwong	*		
Cheilodactylus gibbosus Crested morwong	*		
Mugil cephalus Sea mullet	*		
Etrumeus teres Maray		*	*
Scomber australasicus Blue mackerel		*	*
Hyporamphus sp. Garfish		*	*

Table 11.7: Several fish larvae identified in plankton samples. Type of tow and general time of capture are indicated (* = present)

Taxonomic Group	Tow Type and Date		
	surface, 1000µm July, 1994	vertical, 500µm July, 1994	vertical 500µm July, 1993
Myctophidae	*	*	*
Macrouridae		*	*
Berycidae		*	*
Leptocephalus	*		
Monocanthidae	*	*	*
Callionymidae			*
Cephalopods	*		
Labridae		*	*
Moridae		*	*
Scorpaeniformes	*	*	*

Table 11.8: General taxonomic groups found in the plankton samples. Type of tow and general time of capture are indicated (* = present)

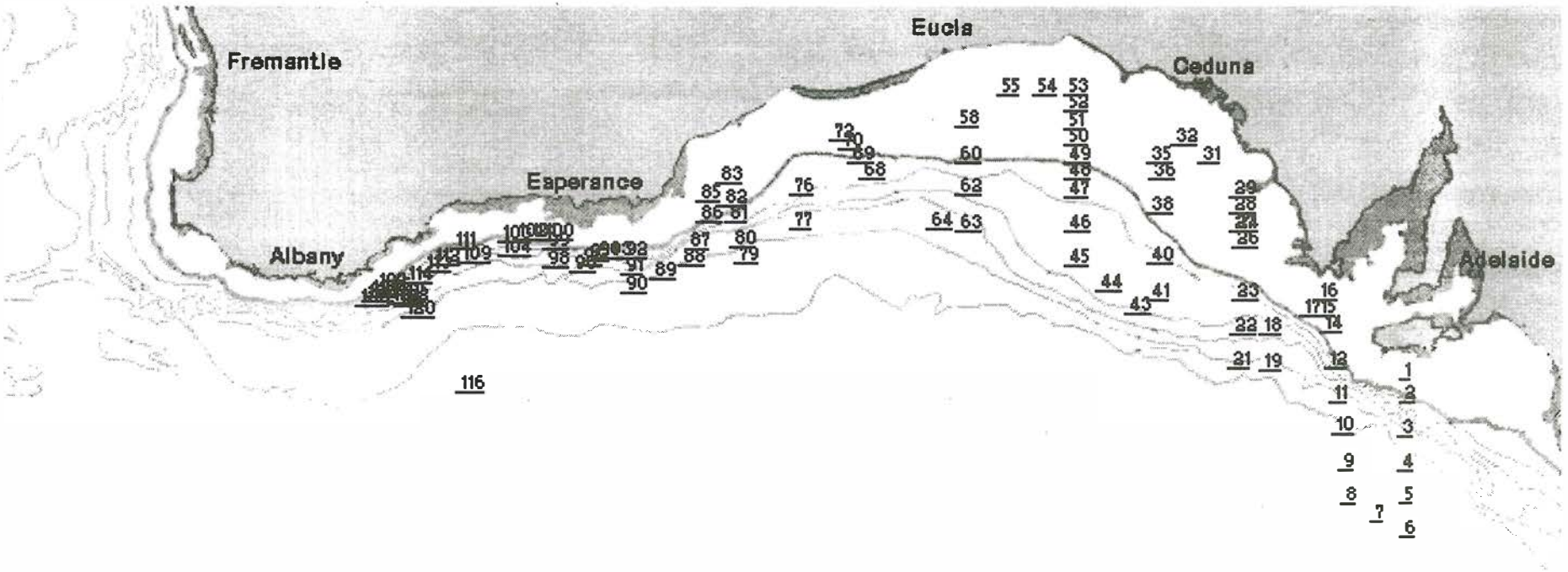

Figure 11.1: Tow number and location of surface tows conducted with 1000 micron mesh for July 1994 plankton survey

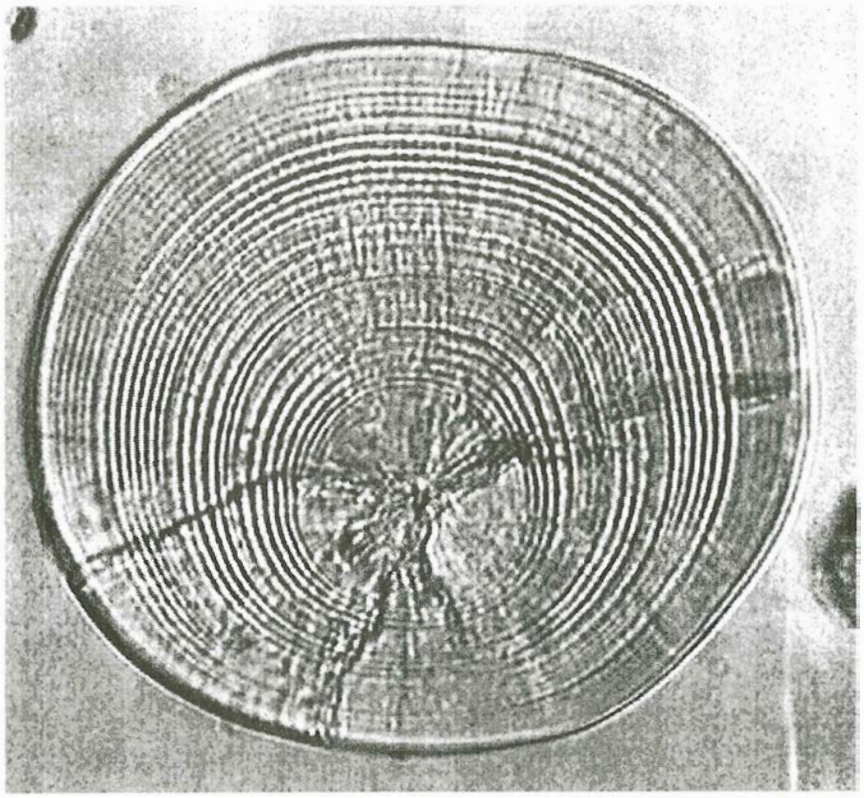

Figure 11.2: Captured image of larval pilchard otolith showing alternating light/dark concentric rings.

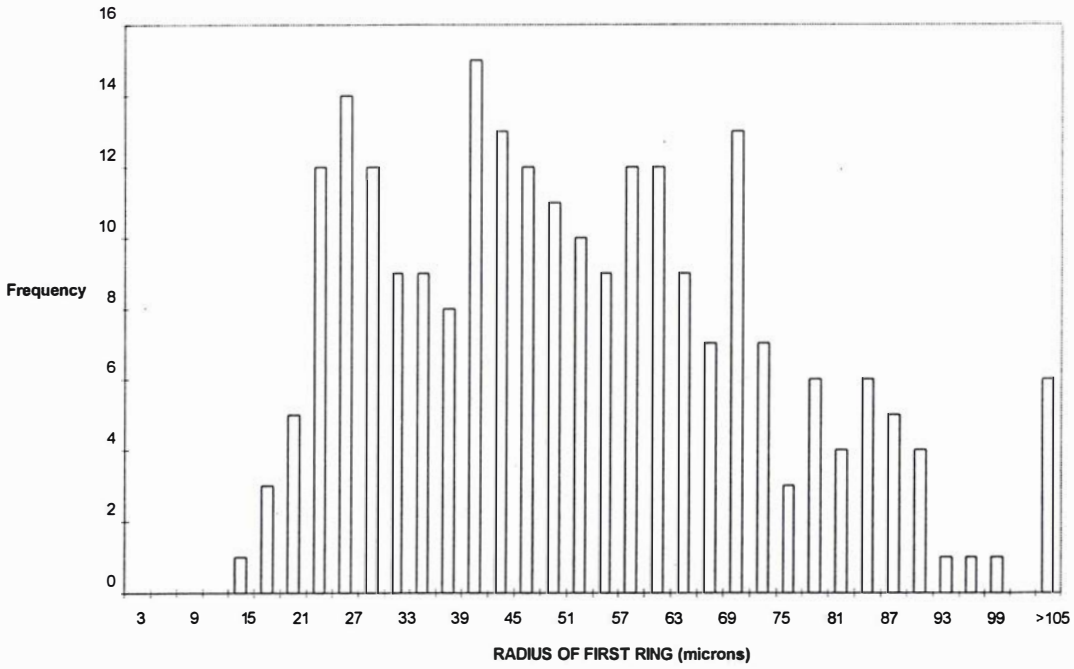

Figure 11.3: Frequency histogram of radius of first ring from otolith focus

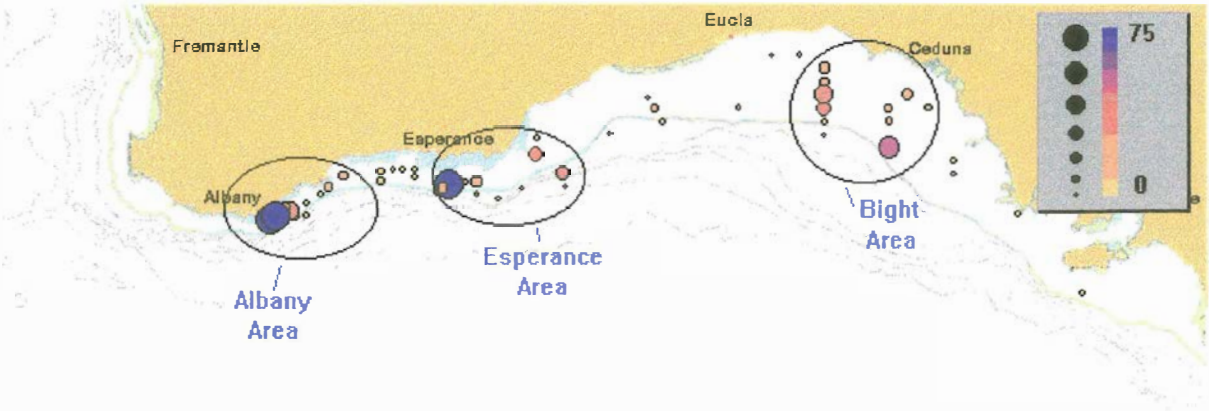

Figure 11.4: Abundance of pilchard larvae found in surface tows conducted with 1000 micron mesh for July 1994 plankton survey . Circles encompass selected areas used in larval growth comparisons.

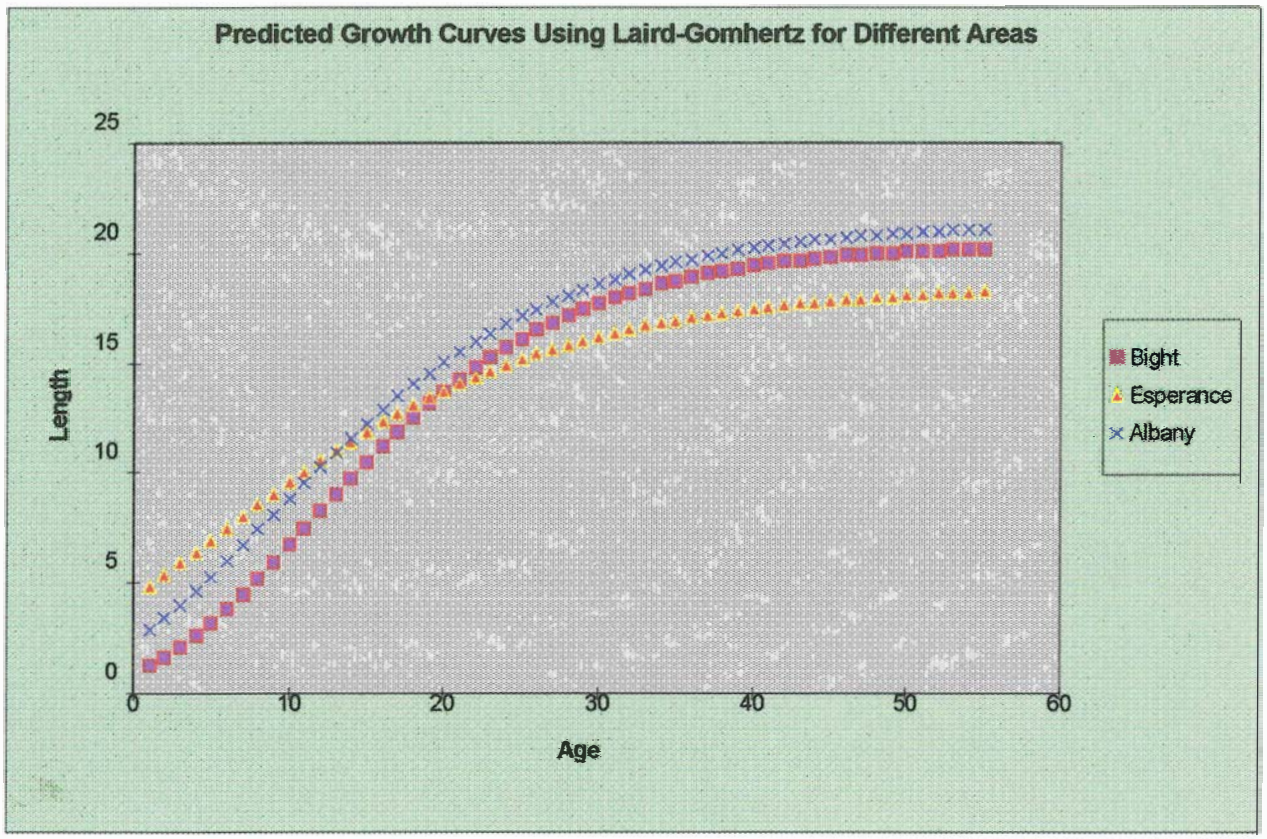

Figure 11.5: Predicted growth curves using the Laird-Gompertz growth function for selected areas along the south coast.

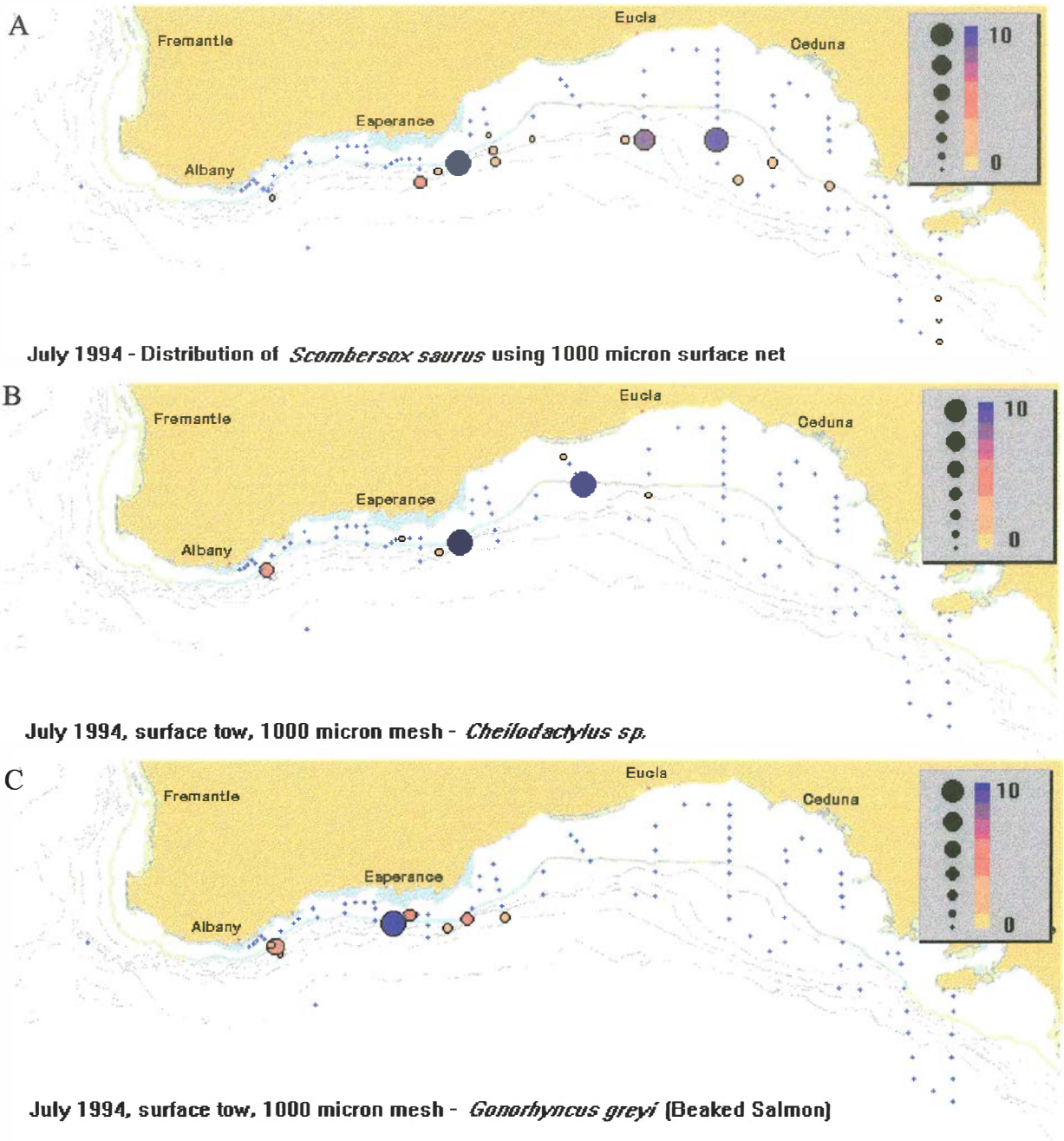

Figure 11.6: Distribution of selected fish larval groups from July 1994 surface tows with 1000 micron mesh (a) *Scombersox saurus* (b) *Cheilodactylus sp.* (c) *Gonorhyncus greyi*

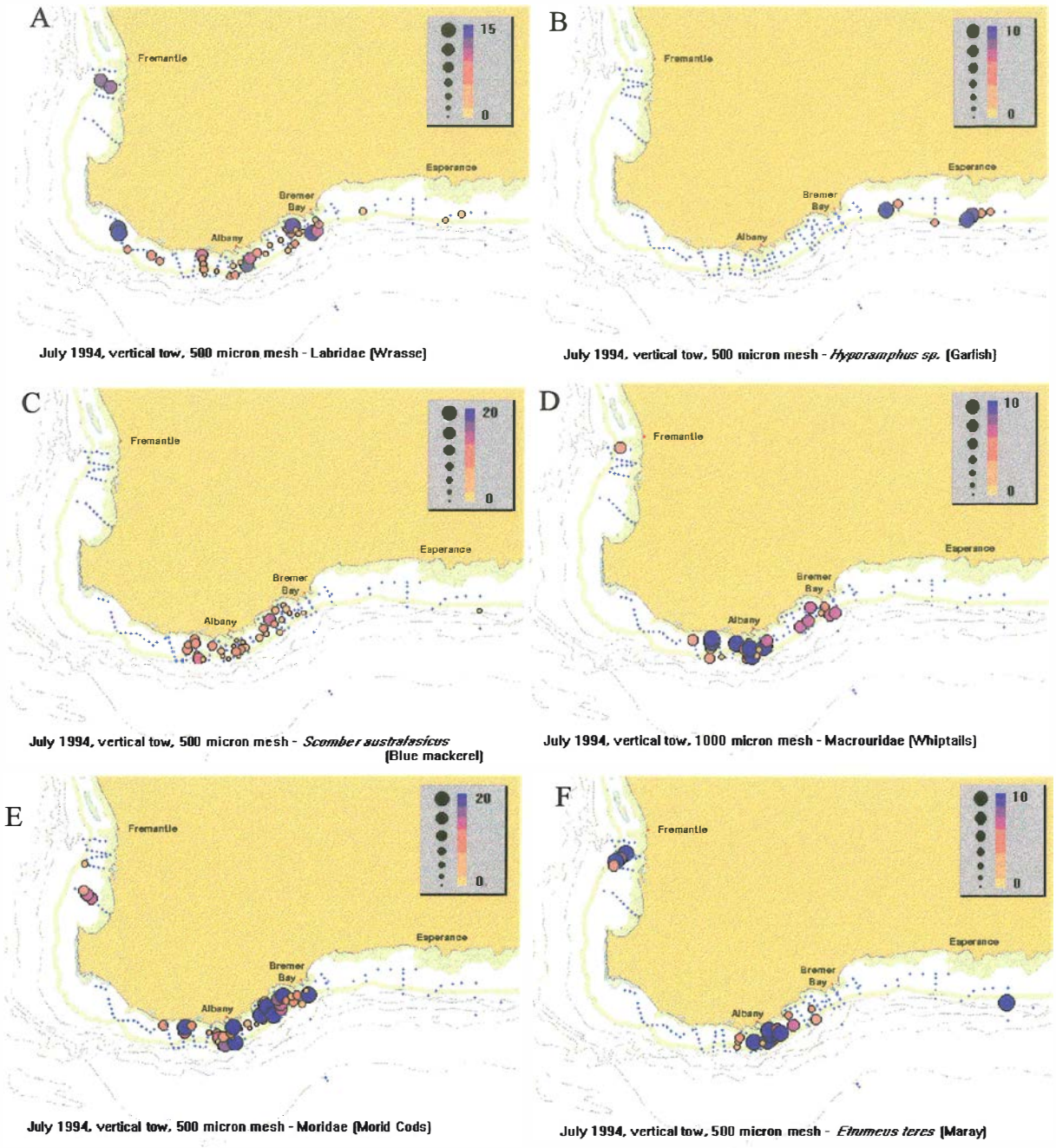

Figure 11.7: Distribution of selected fish larval groups from July 1994 vertical tows with 500 micron mesh (a) Labridae (b) *Hyporamphus sp.* (c) *Scomber australasicus* (d) Macrouridae (e) Moridae (f) *Etrumeus teres*

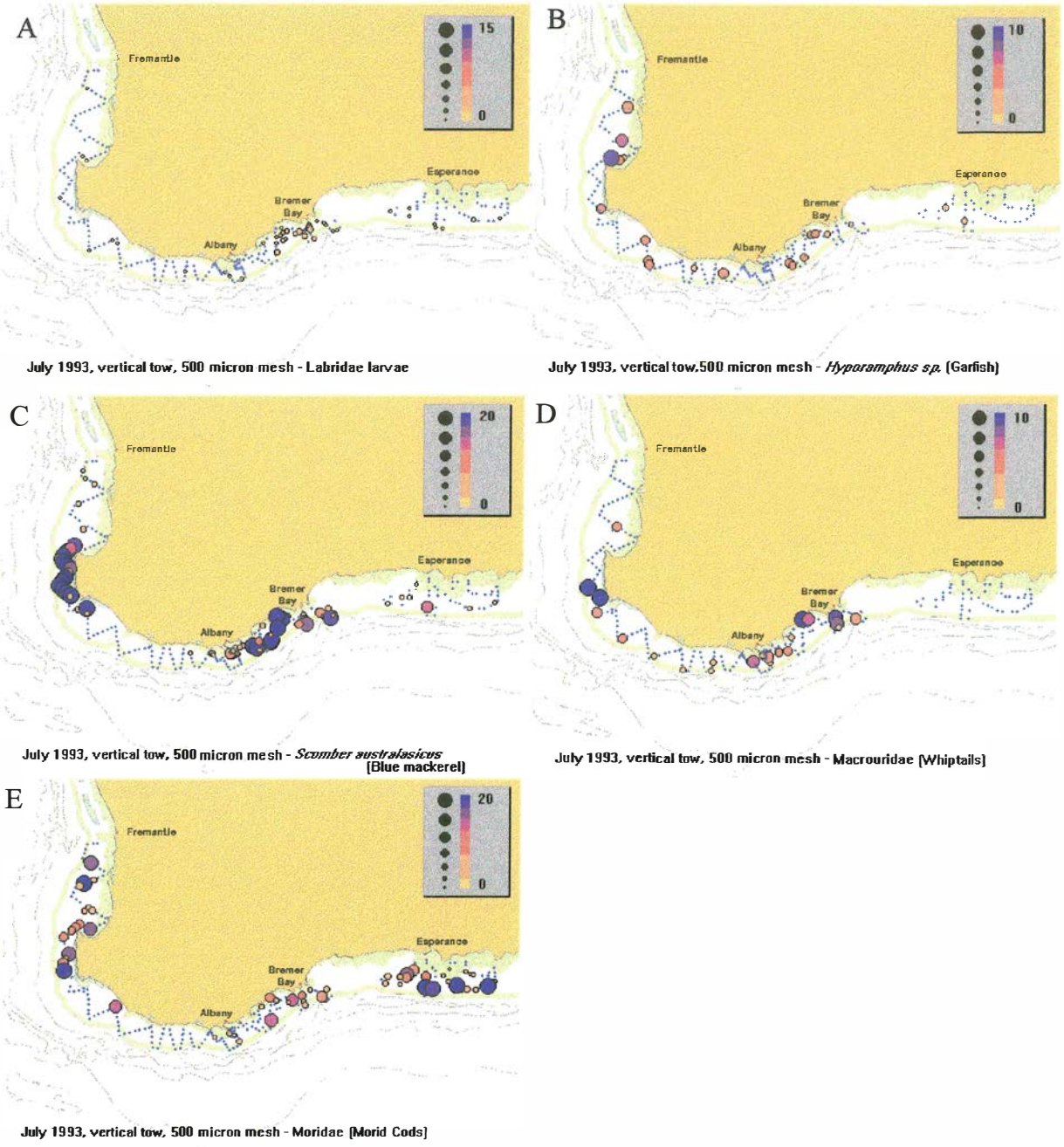

Figure 11.8: Distribution of selected fish larval groups from July 1993 vertical tows with 500 micron mesh (a) Labridae (b) *Hyporamphus sp.* (c) *Scomber australasicus* (d) Macrouridae (e) Moridae

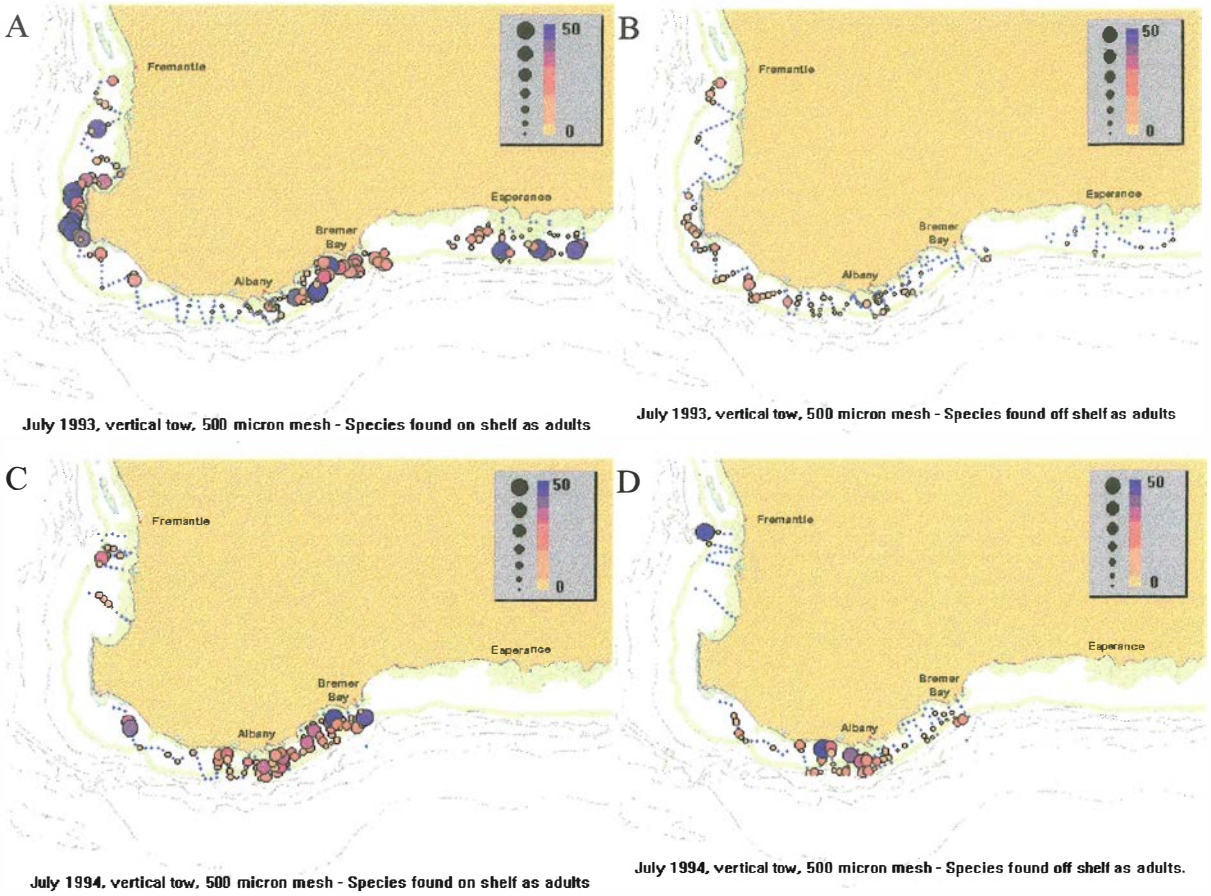

Figure 11.9: Distribution of shallow water compared to deep water species for vertical tows with 500 micron mesh (a) July 1993 - shallow water groups (b) July 1993 - deepwater groups (c) July 1994 - shallow water groups (d) July 1994 - deep water groups

12. Intellectual Property and valuable information

No saleable items were developed during this study.

13. Further Development

No commercial adaptation of these results is possible.

14. Staff

Staff that were employed on the project using FRDC funds were:

Mr K. White, Dr D. Gaughan, Mr G. Sant, Mr S. Field, Mr R. Mijat

Staff who assisted on the project using non-FRDC funds were:

Dr R. Fletcher, Mr S. Blight, Mr R. Tregonning, Mr J. Jenke, Ms L. Hayes, Mr B. Webber, Mr G. Buchanan, Mr C. Dawes-Smith, Mr J. Kelly, Mr J. Chidlow, Ms. N. Tassone

15. Final Cost

Total expenditure from FRDC funds for this project was $ 216 042.52