

Acoustic survey of squid in inshore spawning grounds between Storms River Mouth and Port Alfred from *R.V. Ellen Khuzwayo* and Research Inflatable *Abyss* between 31 October and 20 November 2021

M.A. Soule and I. Hampton

Fisheries Resource Surveys cc.
Cape Town

Executive Summary

This report details acoustic surveys of *Loligo reynaudii* spawning biomass carried out from *R.V. Ellen Khuzwayo* and Research Inflatable *Abyss* within the 50 m isobath between the Storms River mouth and Port Alfred between 31 October and 20 November 2021, within the second annual closed fishing season. The primary frequency on both vessels was 38 kHz. The methods were similar to those used from these vessels in more limited surveys of the area in November 2019 and November 2020. This survey was both longer in time than those surveys, and for the first time covered the entire inshore commercial fishing ground, including the nearshore area shallower than 30 m. In all, 403 lines were worked by *Ellen Khuzwayo* and 185 by *Abyss*. After overlaying the grids the lines were effectively 0.5 n.miles apart, making this the most intensive survey of *L. reynaudii* yet.

The main objective of the survey was to examine the feasibility of obtaining a fisheries-independent index of *L. reynaudii* biomass on its known inshore spawning ground for potential use in managing the resource. The survey was a sequel to two previous acoustic surveys over a narrower geographical range in November 2019 and November 2020 respectively (Soule and Hampton, 2020 and 2021).

Ellen Khuzwayo surveyed between 30 and 50 m in four phases, the first two of which covered the full area, and the latter two the core area, between Aasvögels Bult and Maitlands. With the exception of Algoa Bay, *Abyss* covered this area in water shallower than 30 m to a minimum depth of about 10 m, depending on safety, in two phases, which were synchronised as closely as possible with Phases 1 and 2 of the *Ellen Khuzwayo* surveys. Neither survey was significantly comprised by weather, which was unusually good for most of the time.

Characteristic squid spawning aggregations (classified as A - Category marks) were relatively scarce, with only 11 being detected in total. All of these were located in the core area. Other squid-like, but less characteristic, marks (classified as B - *Probably Squid*) were more widely spread, particularly in the core area. For the first time, weaker, even less characteristic spawning squid marks were also included in the estimate to examine the possibility of a significant proportion of the biomass being concentrated in these marks. They were classified as Category C - *Possibly Squid*).

The biomass and CV estimates from the entire survey, including all phases from both vessels, are summarised in the following Table.

| Vessel | <i>Ellen Khuzwayo</i> | | <i>Abyss</i> | | <i>Ellen Khuzwayo + Abyss</i> | |
|----------|-----------------------|------|------------------|------|-------------------------------|------|
| Category | Biomass (tonnes) | CV | Biomass (tonnes) | CV | Biomass (tonnes) | CV |
| A | 231.5 | 0.51 | 112 | 1.0 | 343.5 | 0.47 |
| B | 322.7 | 0.17 | 260.6 | 0.27 | 583.3 | 0.15 |
| C | 864.6 | 0.15 | 573.7 | 0.25 | 1 438.3 | 0.13 |
| A+B | 554.3 | 0.23 | 260.6 | 0.27 | 926.8 | 0.20 |
| A+B/2 | 392.8 | 0.33 | 130.3 | 0.18 | 635.1 | 0.20 |
| A+B+C | 1 419.1 | 0.13 | 946.4 | 0.20 | 2022 | 0.09 |

We consider the most reliable biomass estimate from these data to be a combination of the A estimate and half the B estimate from the two vessels, which minimises the maximum error incurred from including the B estimates. This estimate is 635 tonnes, with a CV of 0.20 (ie. 20 %). The estimate for the core area (373 tonnes, with a CV of 0.25) is reasonably close to a comparable estimate from *Abyss* for this area in the previous year (viz: 291.8 tonnes; CV 0.21). It is consistent with catches immediately following the survey, which were unusually low, lending further credence to the estimate as an estimate of spawning biomass in the area at the time of the survey. It is also encouraging that estimates of the target strength at 38 kHz of squid, made opportunistically during the survey, are consistent with estimates from our previous studies on *L. reynaudii* target strength at 38 kHz.

The addition of the C - Category marks raised the estimate of total biomass to 2 022 tonnes and reduced the CV to 9 %. However, due to the uncertainty in classifying these marks in the presence of many similar marks from pelagic fish and other species, and the lack of direct evidence at this stage that *L. reynaudii* can indeed form these kinds of mark in abundance on the inshore spawning grounds, we tentatively conclude that this estimate is positively biased, perhaps by a large amount. As such it is probably safe to regard it only as an absolute upper limit, for whatever purpose this may serve at present. Whether it will be worthwhile to include these marks in any future analysis, given that it greatly increases the complexity of the analysis and, because of the large number of marks which have to be scrutinised, the time needed to complete the report, is debateable.

There were sometimes substantial differences in the biomass estimates between the various Phases, but overall, nothing to suggest a large-scale immigration or emigration of the spawning population to or from the survey area during the two-week period of the survey.

We conclude that good progress has been made through this survey in developing a standard acoustic method of monitoring the biomass of spawning *L. reynaudii* in the

November closed season, and that the survey design and methods used here should become the standard for future acoustic surveys of spawning biomass to optimise comparison of the estimates between the years. In any such surveys, it will be important to continue attempting to improve confidence in target recognition, particularly of the weaker targets.

The question of whether the current estimate is a reliable absolute or relative estimate of the size of the entire adult population on the inshore spawning ground, and therefore of its potential use for management of the entire population, is beyond the scope or brief of this study. We suggest that this question be addressed, *inter alia*, through an in-depth examination of the large amount of data available from the commercial fleet on the changes in the location and diurnal catch patterns throughout the year, supplemented by studies of aggregating behaviour outside the November closed season.

Introduction

This document describes an acoustic survey of *Loligo reynaudii* on their inshore spawning grounds between the Storms River mouth and Port Alfred in the closed season between 30 October and 20 November 2021, from Research Vessel *Ellen Khuzwayo* and Research Inflatable *Abyss*. Both vessels were equipped with calibrated Simrad EK60 scientific sounders transmitting simultaneously into 38 and 200 kHz split-beam transducers. *Ellen Khuzwayo* worked between the 30 and 50 m isobaths, and *Abyss* inshore of the 30 m isobath to a minimum depth of approximately 10 m depending on safety considerations. The vessels worked largely independently, but where it was possible the surveys were synchronised to some extent.

The primary objective of the survey was to examine the feasibility of obtaining a fisheries-independent index of *L. reynaudii* biomass on its known inshore spawning ground for potential use in managing the resource. The survey was a sequel to two previous acoustic surveys over a narrower geographical range in November 2019 and November 2020 respectively (Soule and Hampton, 2020 and 2021).

Personnel

Ellen Khuzwayo

Mike Soule (Fisheries Resource Surveys - FRS)

Jean Waruguru Mwicigi (Department of Forestry, Fisheries and the Environment – DFFE)

Abyss (variously)

Johan Rademan (DFFE)

Corne Erasmus (DFFE)

Dagmar Merkle (DFFE)

Johan Mhlongo (DFFE)

Rob Cooper (Bayworld Centre for Research and Education – BCRE)

Ashore, Cape St Francis

Ian Hampton (FRS)

Vessels and Equipment

R.V. Ellen Khuzwayo (Fig. 1) is a 43.2 – m research vessel equipped with a multi-frequency Simrad EK60 scientific echo sounder firing into 38, 120 and 200 kHz split-beam transducers co-located on the hull. *Abyss* (Fig. 2) is a 7.5 - m rigid-hulled research inflatable, also fitted with a multi-frequency Simrad EK60 sounder, in this case firing into 38 and 200 kHz split-beam transducers co-located on a housing which could be lowered to about 0.5 m below the surface on a retractable pole. Both vessels are owned by DFFE.



Fig. 1: *R.V. Ellen Khuzwayo*



Fig. 2: Research Inflatable *Abyss*

Specifications of the systems used on this survey, and the operational settings, are listed in Table 1. Note that the calibration on *Ellen Khuzwayo* was done immediately before the start of the survey, while *Abyss* was first calibrated on 16 April 2021, then again some six months after the survey, on 17 May 2022. The report on the *Ellen Khuzwayo* calibration is appended

as Annexure A, and that of both *Abyss* calibrations, in Appendix B. Conditions were good and the RMS error low for all of these calibrations (see Appendices A and B). The good agreement of the two calibrations in Appendix B indicate that the system on *Abyss* has been stable over the past year.

Raw acoustic data were logged via the SIMRAD EK60 echo sounders during surveys and drifts and analysed ashore through EchoView Version 5.3 (*Ellen Khuzwayo*) and Version 12.1 (*Abyss*).

| Vessel | Survey dates | Frequencies (kHz) | Nominal -3 dB beamwidth (deg) | Pulse duration (ms) | Calibrations |
|-----------------------|--------------|-------------------|-------------------------------|---------------------|--------------|
| <i>Ellen Khuzwayo</i> | 6/11 – 20/11 | 38 | 7.0 | 0.512 | 5/11/2021 |
| | | 200 | 7.0 | 0.512 | |
| <i>Abyss</i> | 31/10- 15/11 | 38 | 12.0 | 0.512 | 16/4/2021 |
| | | 200 | 7.0 | 0.128 | 17/5/2022 |

Table 1: Dates of surveys by *R.V. Ellen Khuzwayo* and Research Inflatable *Abyss* in November 2021, and details of Simrad EK60 echo sounders used on each.

Surveys

Based largely on the topography of the coastline, the survey was divided into four main strata, defined as follows:

- Stratum 1: Storms River - Cape Seal
- Stratum 2: Cape Seal – Cape Recife
- Stratum 3: Algoa Bay (Cape Recife) – Woody Cape
- Stratum 4: Woody Cape – Port Alfred

These strata were further divided according to differences in expected squid densities within them into the strata listed in Table 2. The Table also gives the dates during which the strata were surveyed by *Ellen Khuzwayo*, and the number of transects per stratum. The survey speed was set at 10 knots throughout (occasionally slightly higher). The line spacing was fixed at 1.0 n.miles. *Ellen Khuzwayo* surveyed the area in four consecutive phases, the first two of which covered the whole area, and the latter two parts of Stratum 1 and parts of Stratum 2, as shown in Table 2. Note that surveys of the strata with the same number are nominally replicates. To keep the average time separation between the Phase 1 and Phase 2 transects approximately constant at six days, the vessel returned to Storms River after completion of the Phase 1 survey, to start Phase 2. Note that the lines in Phase 2 were interleaved with those in Phase 1, effectively creating a spacing of 0.5 n.miles for the two surveys combined. The lines in Phase 3 duplicated those in Phase 1, and those in Phase 4 the lines in Phase 2, again creating a net spacing of 0.5 n.miles for the two surveys combined. All surveying was done in daylight hours. The total distance steamed in each of the strata in Table 2 was limited to allow the surveys to be completed comfortably between 06h00 and 18h00, except for Algoa Bay (Stratum 3), which took two days to cover.

| Phase | Stratum | Date (Nov) | Alongshore limits | No. lines |
|------------|---------|------------|-------------------------------|-----------|
| 1 | 1B | 6 | Aasvögels Bult – Cape Seal | 28 |
| | 2A | 7 | Cape Seal –Jeffreys Bay | 14 |
| | 2B | 7 | Jeffreys Bay – Maitlands | 14 |
| | 3 | 8 - 9 | Cape Recife – Woody Cape | 34 |
| | 4 | 10 | Woody Cape - Port Alfred | 34 |
| 2 | 1A | 11 | Storms River – Aasvögels Bult | 22 |
| | 1B | 12 | Aasvögels Bult – Cape Seal | 28 |
| | 2A | 13 | Cape Seal – Jeffreys Bay | 14 |
| | 2B | 13 | Jeffreys Bay - Maitlands | 14 |
| | 2C | 14 | Maitlands - Cape Recife | 21 |
| | 3 | 14 -16 | Cape Recife – Woody Cape | 35 |
| | 4 | 16 | Woody Cape – Port Alfred | 33 |
| 3 | 1B | 19 | Aasvögels Bult - Cape Seal | 28 |
| | 2A | 17 | Cape Seal – Jeffreys Bay | 14 |
| | 2B | 17 | Jeffreys Bay -Maitlands | 14 |
| 4 | 1B | 20 | Aasvögels Bult -Cape Seal | 28 |
| | 2A | 18 | Cape Seal – Jeffreys Bay | 14 |
| | 2B | 18 | Jeffreys Bay - Maitlands | 14 |
| All strata | | | | 403 |

Table 2: Details of grids worked in survey by *Ellen Khuzwayo*. Note that surveys of strata with the same number are nominally replicates.

Except for Stratum 3 (Algoa Bay), which she did not survey, *Abyss* surveyed the same strata as *Ellen Khuzwayo* in Phase 1, with the addition of Stratum 1A (Storms River to Aasvögels Bult) and 2C (Maitlands - Cape Recife) which was not surveyed by *Ellen Khuzwayo* in her Phase 1 survey. In Phase 2, *Abyss* conducted surveys between Aasvögels Bult and Maitlands (ie. Strata 1B, 2A and 2B). Further details of the surveys, which were all run at around 6 knots, on a fixed line spacing of 1.0 n.miles, are given in Table 3. The lines in the Phase 2 survey were also offset by 0.5 n.miles in relation to those in Phase 1, creating an effective line spacing of 0.5 n.miles for these two surveys combined as well.

| Phase | Stratum | Date | Alongshore limits | No. lines |
|------------|---------|-------|-------------------------------|-----------|
| 1 | 1A | 31/10 | Storms River – Aasvögels Bult | 22 |
| | 1B | 3/11 | Aasvögels Bult - Cape Seal | 28 |
| | 2A | 4/11 | Cape Seal - Jeffreys Bay | 14 |
| | 2B | 6/11 | Jeffreys Bay – Maitlands | 16 |
| | 2C | 7/11 | Maitlands – Cape Recife | 21 |
| | 4 | 10/11 | Woody Cape - Port Alfred | 26 |
| 2 | 1B | 12/11 | Aasvögels Bult – Cape Seal | 28 |
| | 2A | 14/11 | Cape Seal – Jeffreys Bay | 15 |
| | 2B | 15/11 | Jeffreys Bay - Maitlands | 15 |
| All strata | | | | 185 |

Table 3: Details of wide area grids worked in survey by *Abyss*. Note that surveys of strata with the same number are nominally replicates.

The grids of the Phase 1 and Phase 2 surveys by *Ellen Khuzwayo* and *Abyss* are shown in Figs. 3a and 3b respectively, and those of Phases 3 and 4 by *Ellen Khuzwayo*, in Fig. 4.

In addition to the formal surveys, *Ellen Khuzwayo* conducted one *ad hoc* intensive survey of an A - Category mark at Jeffreys on 17 November on a star grid to collect further information on the target strength of *L. reynaudii* at 38 and 200 kHz. The purpose of this was to search for differences in their target strength at these two frequencies as a potential target-identification tool, and to add to the pool of target strength data at 38 kHz collected in previous work, as summarised in Soule *et al.* (2010). Target strength data were also collected by *Abyss* on a parallel grid survey of A - Category marks and drifts at Jeffreys on 4, 6 and 14 November, and the Kromme on 15 November. The positions of these experiments are indicated on Fig. 5. Note that target strength estimates at 38 kHz from these experiments were not used directly in the biomass calculations, since the expression which was used (Eqn. 1) is better founded, being based on a much broader set of both *in situ* and *ex situ* experiments over a much longer time period. Rather, they were used to expand the earlier data set and, it was hoped, to increase confidence in the earlier results.

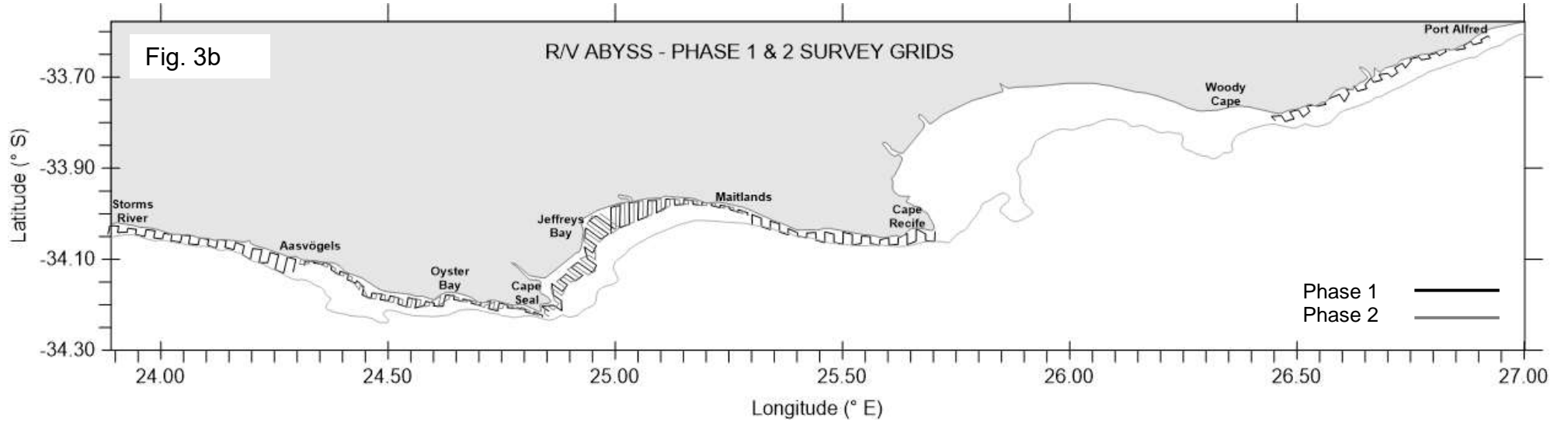
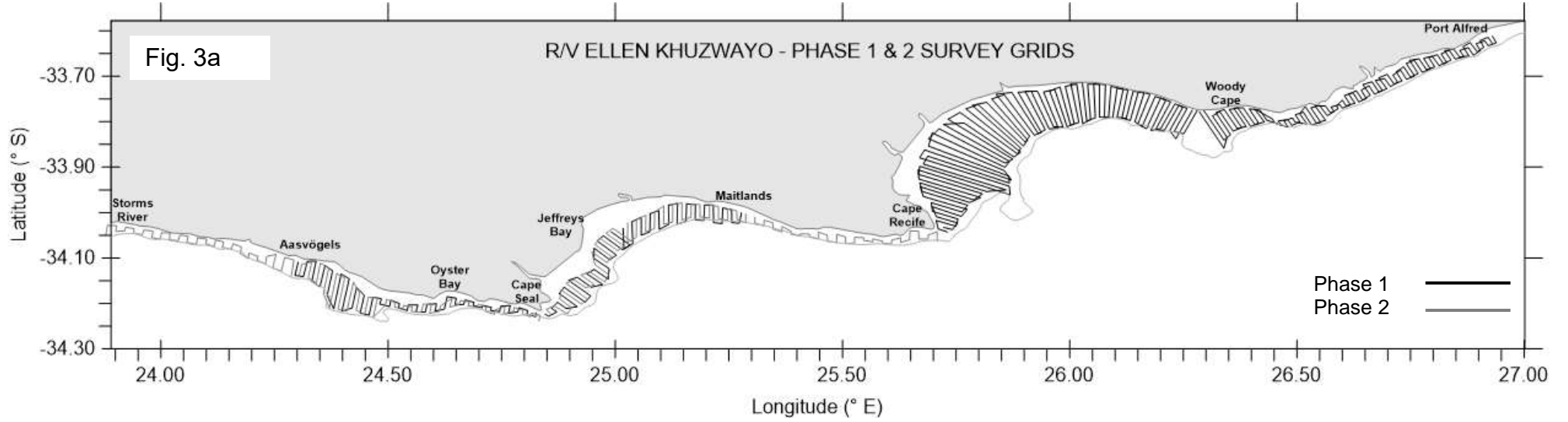


Fig. 3: Grids worked by *Ellen Khuzwayo* (3a) and *Abyss* (3b) in Phases 1 (black) and 2 (grey).

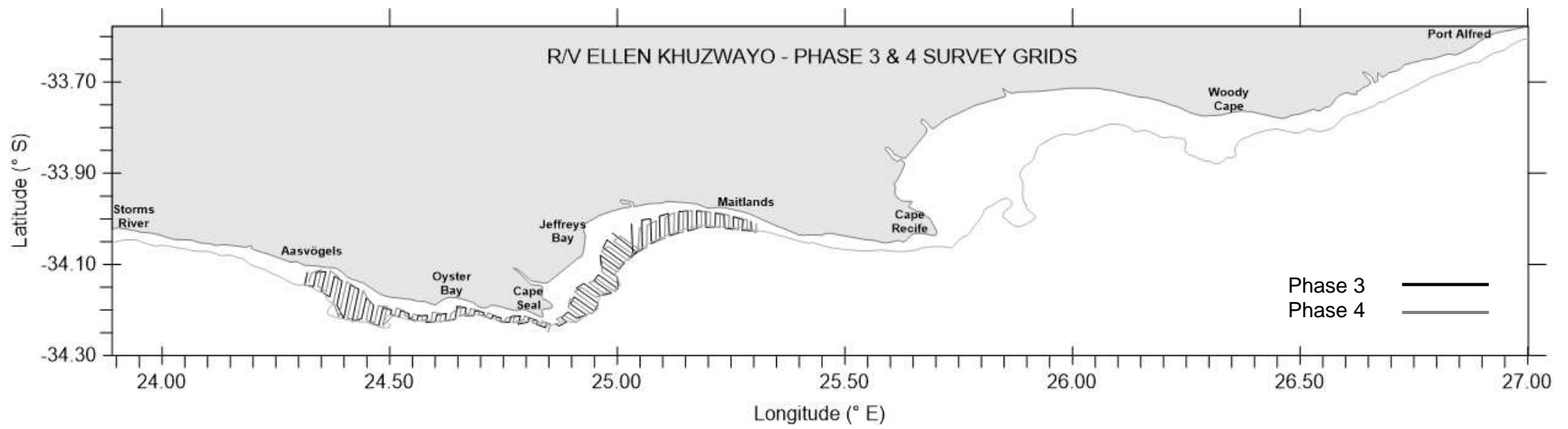


Fig. 4: Grids worked by *Ellen Khuzwayo* in Phases 3 (black) and 4 (grey)

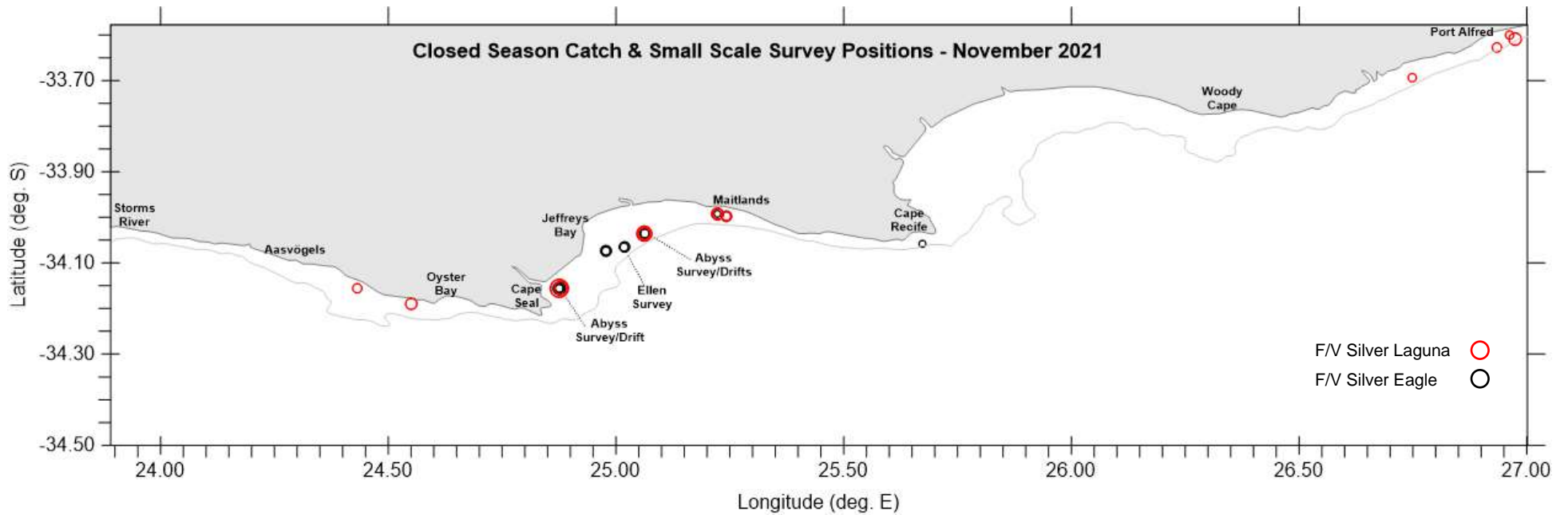


Fig. 5: Positions of the *ad hoc* small-scale surveys of A - Category marks by *Ellen Khuzwayo* and *Abyss*, and of the catches by *F.V. Silver Eagle* (black) and *F.V. Silver Laguna* (red) used in the analysis. The size of the circle roughly represents catch size.

Biological data on squid in the survey area were collected by jig from two commercial vessels engaged to work under the direction of *Ellen Khuzwayo*. The first of these (*F.V. Silver Eagle*; Skipper Craig van Rij) operated primarily between Aasvögels and Cape Recife, and the second (*F.V. Silver Laguna*; Skipper Marius Breytenbach), between Cape Seal and Port Alfred. The positions of the catches used in the analysis to provide length and weight information are shown in Fig. 5.

Data Analysis

Target classification

A key aspect of the analysis was deciding whether marks detected on the transects were squid or not. In the 2019 and 2020 surveys (Soule and Hampton, 2020, Soule and Hampton, 2021) marks thought to be from squid were classified into two categories: an A category where the marks were highly characteristic of spawning aggregations, often having a familiar “mushroom” shape such as that shown in Fig. 6, and a B category where the aggregations were of similar density to the A marks, and sometimes in close proximity to them, but had less distinctive conformations (see example in Fig. 6). These marks were regarded as “*probably squid*” although with less certainty than the A marks. In some cases, where the density was low enough for the resolution of echoes from many individuals, support for the classification as squid was derived from the presence of a significant peak in the target strength distribution between -40 and -45 dB, where most of the target strength values in the study by Soule *et al.* (2010) on *L. reynaudii* lay.

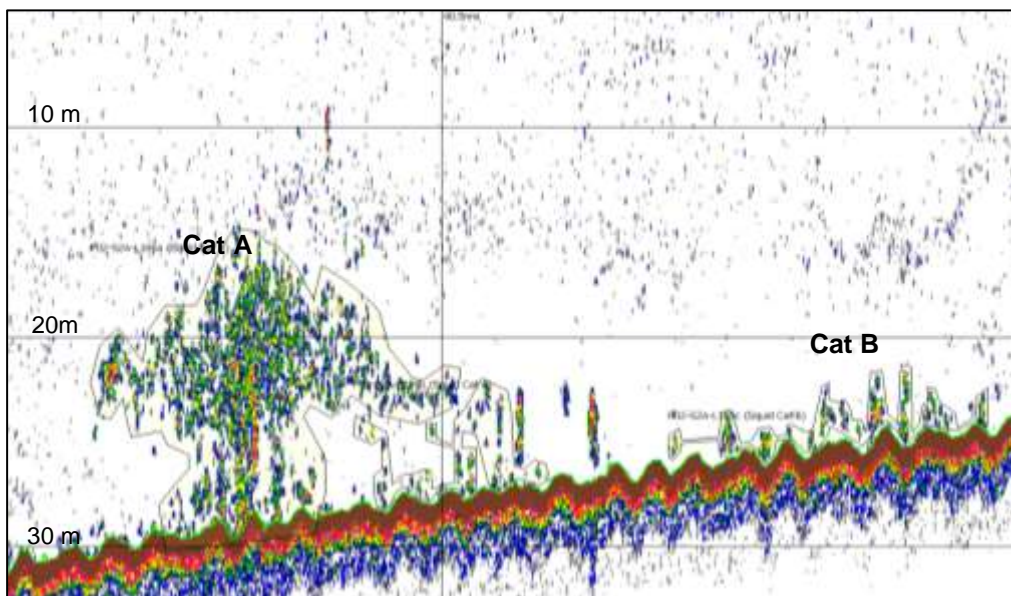


Fig. 6: Example of a “mushroom-shaped” Cat. A mark and nearby less-distinctive marks placed in the B Category (*probably squid*) recorded from *Ellen Khuzwayo* while surveying Line 165 off Jeffreys Bay during Phase 2 on 13 November..(34° 03.906' S, 25° 01.158' E).

In the current survey, a third category (Category C – *possibly squid*) was added to explore the possibility of a significant proportion of the biomass being concentrated in small, often scattered aggregations of low density in the lower half of the water column by day, mixed with other targets such as small pelagic fish schools (particularly of juvenile round herring

Etrumeus whiteheadi, juvenile horse mackerel *Trachurus trachurus capensis* and anchovy *Engraulis capensis* which are abundant in the area.¹ Examples are shown in Figs. 7a and 7b.

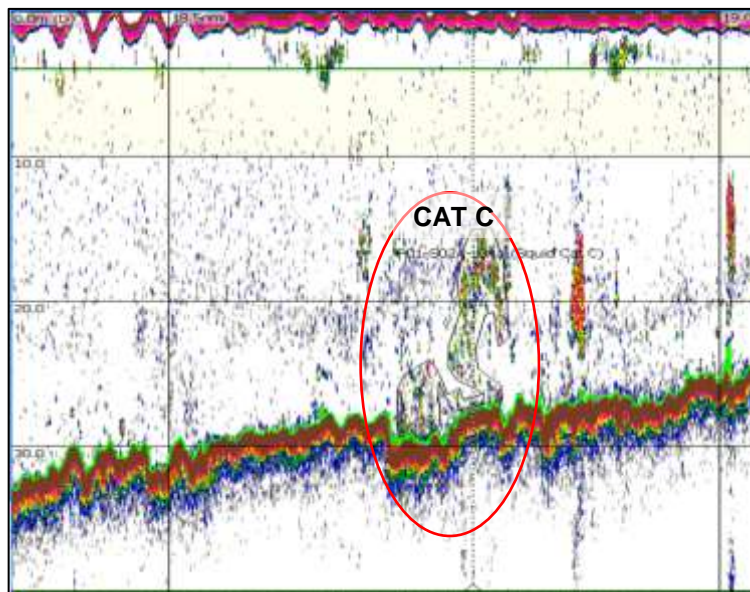


Fig.7a: Example of marks classified as “possibly squid” (Category C) in the vicinity of much denser pelagic fish schools, probably of round herring, juvenile horse mackerel and/or anchovy, recorded from *Ellen Khuzwayo* while surveying Line 34 off the Kromme during Phase 1 on 7 November.

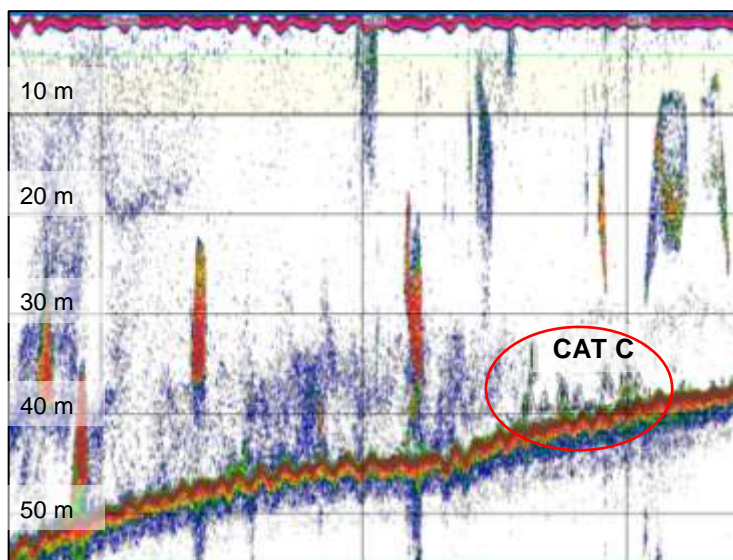


Fig.7b: Example of marks classified as “possibly squid” (Category C) on the bottom in the vicinity of a broad scattering layer in midwater, possibly from pelagic fish. Recorded on *Ellen Khuzwayo* Line 54 off Blue Horizon Bay (Stratum 2B) during Phase 1 on 7 November.

¹ In a small study from *Abyss* six months later using lures (Soule, 2022), it was shown that there are many sparids and other species in the area (some of which form schools) which return echoes similar to some of those we classified as C - Category squid marks. A summary report is appended as Appendix D.

The identification and classification of these targets was far less certain than that of the A and B - Category marks. Consequently they were included in the analysis primarily as a sensitivity study to assess the possible error in the survey estimates due to squid being in aggregations or scattered formations which are more difficult than A and B marks to identify and separate from fish marks. For a mark to be accepted as a C - Category squid target it had to satisfy the following criteria:

- Be in the lower half of the water column, close to, or on the bottom,
- Have a definable boundary, usually with a broken (non-compact) internal density structure,
- Have a maximum volume back-scattering strength less than about -33 dB, which equates to a volume density of the order of 10 animals m⁻³, based on target strength values at 38 kHz in Soule *et al.* (2010). This is in contrast to densities at least an order of magnitude higher expected in pelagic fish schools,
- Have significant values between -40 and -45 dB in the target strength distribution- viz. Soule *et al.* (2010).

It is appreciated that in many cases the C - Category classifications are somewhat arbitrary, and that the probably of mis-classifications is high. There is also the possibility of squid targets dispersed on the bottom not being detected at all because of interference from the bottom echo and other dead-zone issues (eg. Ona and Mitson, 1996), or not being sufficiently aggregated to be classified as squid according to any of the above criteria. Both would lead to underestimates in biomass.

Biomass estimation

Biomass estimates were made from marks in the A, B and C Categories using standard echo-integration theory (cf. Simmonds and McLennan, 2005), and following procedures employed in the analysis of the acoustic data from the two previous acoustic surveys of squid in this area as reported by Soule and Hampton (2020) and Soule and Hampton (2021). As in those surveys, target strength estimates were made by applying the 38 kHz target strength/ length expression of Soule *et al.* (2010), viz.

$$TS = 15.99 \text{ Log } ML - 65.80 \quad , \quad (1)$$

where ML is the mantle length in cm, to a pooled length distribution derived from all samples taken from the catches by the commercial vessels where a significant number of females (arbitrarily taken as > 5) were caught. Because of the known bias towards males in jig catches (Lipinski, 1994), the separate pooled length frequencies for males and females were given equal weight when averaged, on the assumption that the true male/female ratio in the population is close to 1.

The mean back-scattering strength per stratum was estimated from the weighted (by transect length) mean of the back-scattering strength per transect for the aggregations along the transect, and the CV from the variation between the transect means. The relevant expressions were derived from Jolly and Hampton's (1990) expressions for randomly-

spaced parallel transects of unequal length. Expressed in terms of the mean areal back-scattering coefficient, S_a , these expressions are:

$$\bar{S}_a = \frac{\sum_{i=1}^n (\bar{S}_a)_i L_i}{\sum_{i=1}^n L_i} \quad (2)$$

$$Var(\bar{S}_a) = \frac{n}{(n-1)} \frac{\sum_{i=1}^n L_i^2 [(\bar{S}_a)_i - \bar{S}_a]^2}{(\sum_{i=1}^n L_i)^2}, \quad (3)$$

where L_i is the length of transect i , $(S_a)_i$ is the S_a for transect i and n the number of transects in the stratum.

Mean densities and CVs for each stratum and phase were applied to estimates of stratum area to give estimates of squid biomass and CV for each stratum and phase. These estimates were combined in various ways (see following Section) to give biomass and CV estimates for various regions, both vessels, and for all three identification categories. When combining estimates for more than one stratum, they were averaged if the strata covered the same area (ie. were nominally replicates), but were added if they did not.

Results

Length distributions

Pooled length distributions of male and female squid aggregated from all catches made by the commercial vessels during the survey are plotted in Fig. 8.

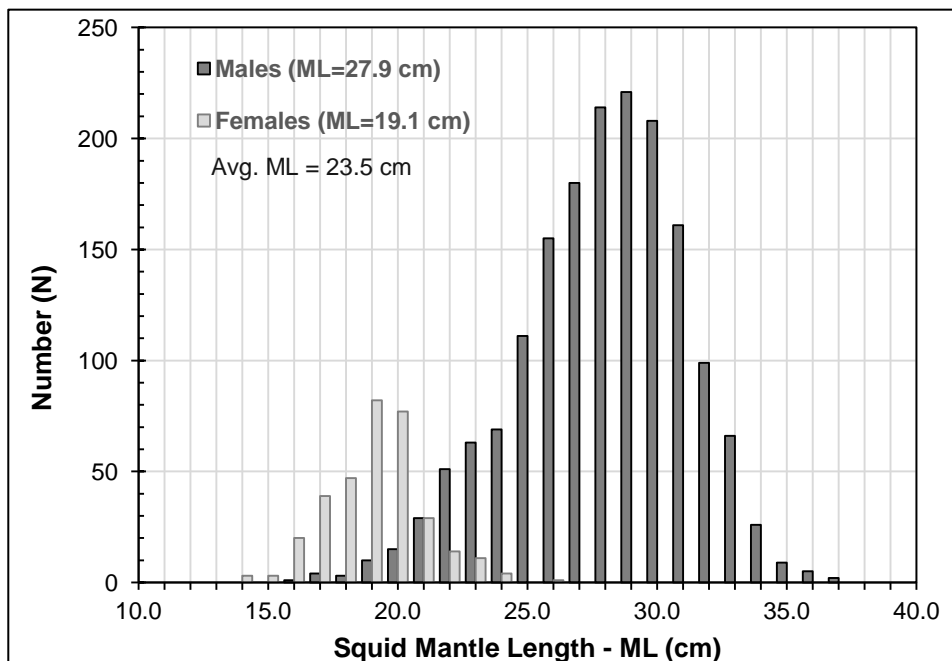


Fig. 8: Pooled length distributions for male and female squid for all squid catches sampled on *F/V Silver Eagle* ($n=14$) and *Silver Laguna* ($n=14$) between Storms River Mouth and Port Alfred from 3 to 15 November. Also shown are the mean length and weight of males and females, and the average of the two (Avg. ML).

Note that no weighting has been applied for the difference in the number of males and females caught for reasons explained in the previous section. The data on which Fig. 8 is based are given in Appendix C. Also shown in the Appendix are the mean mantle length and weight of males and females combined.

Target strength estimates

Ten sets of data were collected simultaneously at 38 and 200 kHz from the A - Category mark surveyed on the star grid by *Ellen Khuzwayo* on 17 November off Jeffreys Bay. The five most comprehensive of these were selected for analysis following protocols established by Soule *et al.* (2010). For each set the number of echoes from what were apparently individual squid, and the mean target strength for both frequencies, are shown in Table 4. A pooled target strength – length distribution constructed from these data without truncation at the upper or lower end is shown in Fig. 9. Note that in deriving the estimates in Table 4 from the individual distributions, they were truncated at -37 dB at the upper end to reduce bias from extraneous strong targets and multiple echoes from squid.

| Experiment | 38 kHz | | 200 kHz | |
|--------------|--------------------------|--------------|--------------------------|--------------|
| | No. of individual echoes | Mean TS (dB) | No. of individual echoes | Mean TS (dB) |
| TS01 | 136 | -43.4 | 105 | -42.7 |
| TS02 | 19 | -40.5 | 26 | -40.4 |
| TS03 | 55 | -43.2 | 53 | -43.0 |
| TS04 | 131 | -43.6 | 116 | -42.2 |
| TS05 | 78 | -41.9 | 83 | -42.7 |
| Mean TS (dB) | | -42.4 | | -42.1 |

Table 4: Estimates of *L. reynaudii* target strength at 38 and 200 kHz from the star survey by *Ellen Khuzwayo* of an A - Category mark off Jeffreys Bay on 17 November. The estimated mean mantle length in the population was 23.5 cm and the estimated mean weight, based on all catches, 0.22 kg (see Fig. 8). All target strength estimates were averaged in the arithmetic domain. Targets were only accepted as single if close to the acoustic axis (maximum beam compensation; 6dB). The distributions were truncated at -37 dB at the upper end to reduce bias from extraneous strong targets and multiple echoes from squid.

These results show no consistent difference in the target strength at 38 and 200 kHz which might be used to discriminate between squid and other targets such as pelagic fish, which also commonly show no consistent difference in back-scattering at these two frequencies (pers. comm. Janet Coetzee, DFFE).

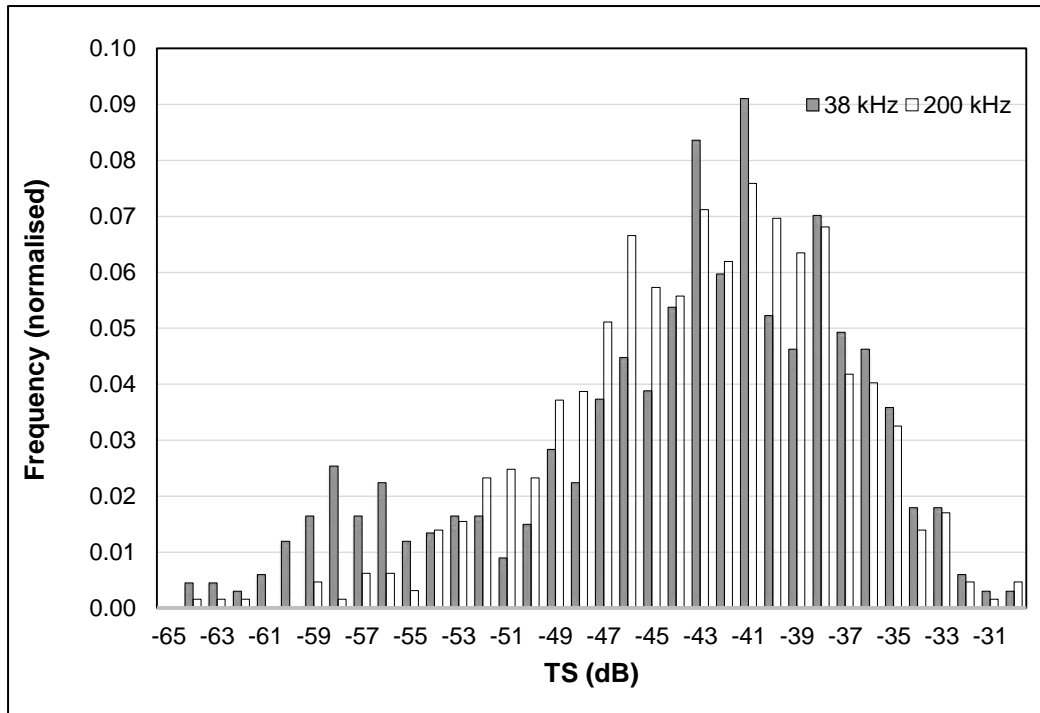


Fig. 9: Target strength distributions at 38 and 200 kHz extracted from data recorded while conducting a star survey over spawning squid off Jeffreys Bay on 17 November.

We note that the mean TS per kg, defined as

$$TS_{kg} = TS_{ind} - 10 \text{ Log } W_{ind} \quad , \quad (4)$$

where TS_{ind} and W_{ind} are the mean target strength and weight per individual, is -35.8 dB at 38 kHz, and -42.2 dB at 200 kHz. Both estimates are close to the means of -36.07 dB kg⁻¹ and 35.64 dB kg⁻¹ in Table 7 of Soule *et al.* (2010) for their Truncation and Gaussian methods respectively of filtering TS distributions from raw data. However, use of the mean length of 23.5 cm in Eqn. 1 gives a value of -43.9 dB for TS_{ind} which differs by 1.5 dB from the 38 kHz value in Table 4.

Distribution

Figs. 10a and 10b show the presence of the A, B and C – Category marks in Phases 1 and 2 of the *Ellen Khuzwayo* survey, and Figs. 11a and 11b the presence of these marks in Phases 3 and 4 of the survey. Figs. 12a and 12b show their distribution in Phases 1 and 2 of the *Abyss* survey.

Biomass estimates

Tables 5 and 6 show the estimates of biomass and associated CV for A, B and C - Category marks for all strata in the *Ellen Khuzwayo* and *Abyss* surveys respectively.

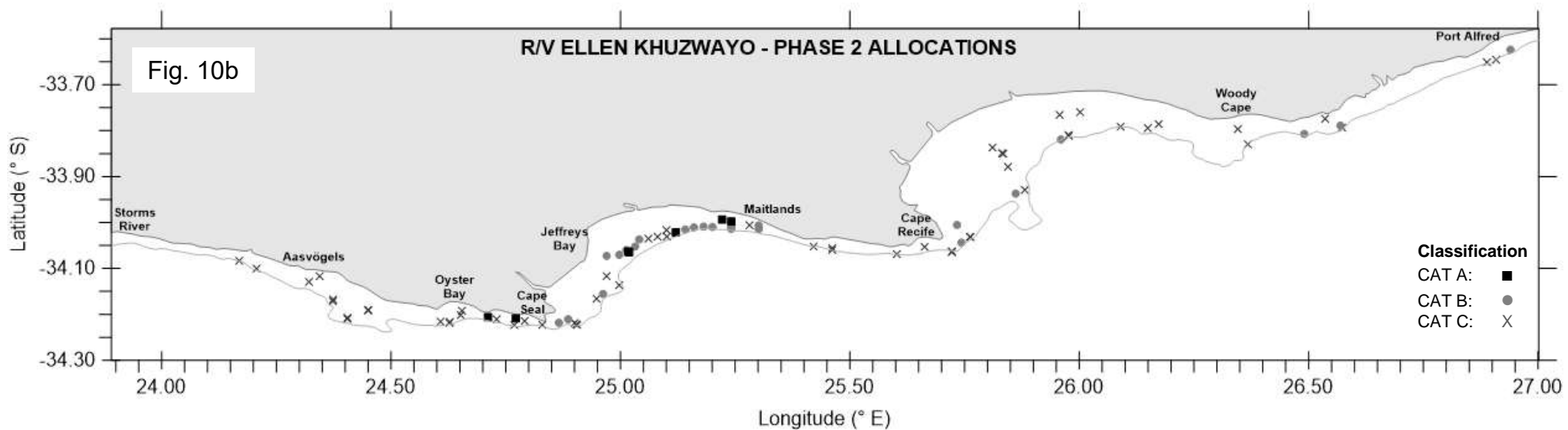
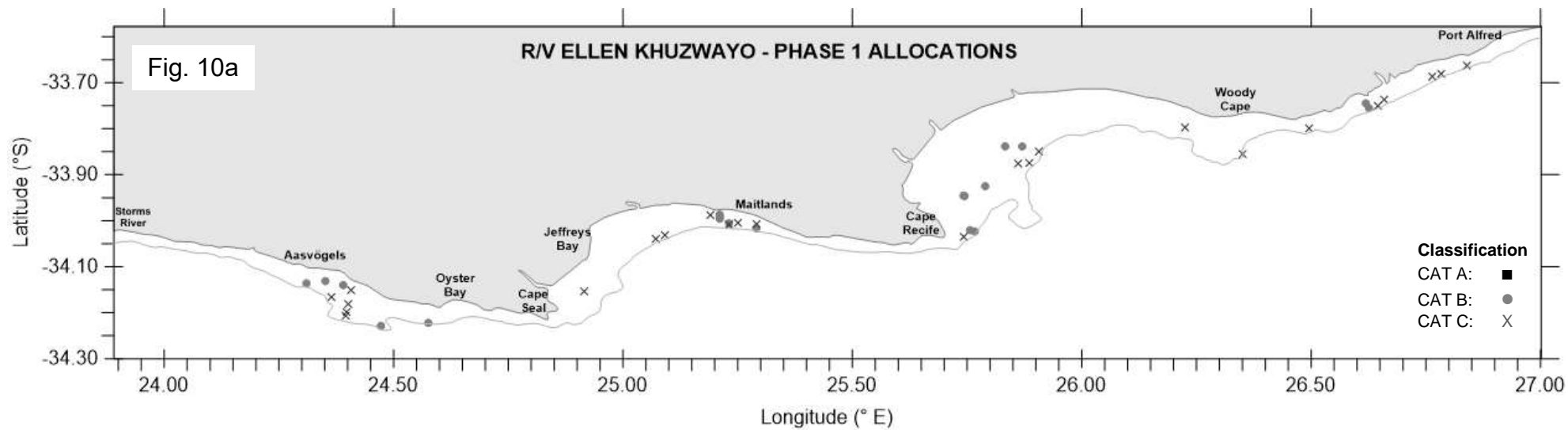


Fig. 10: Location of the A, B and C- Category marks in Phase 1 (Fig. 10a) and Phase 2 (Fig. 10b) of the *Ellen Khuzwayo* survey.

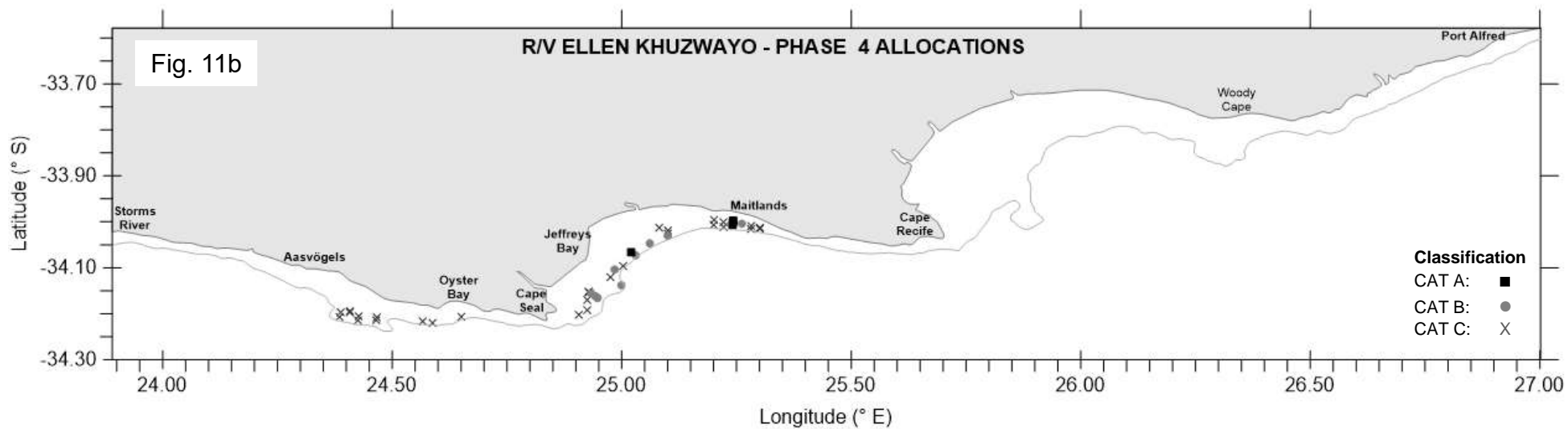
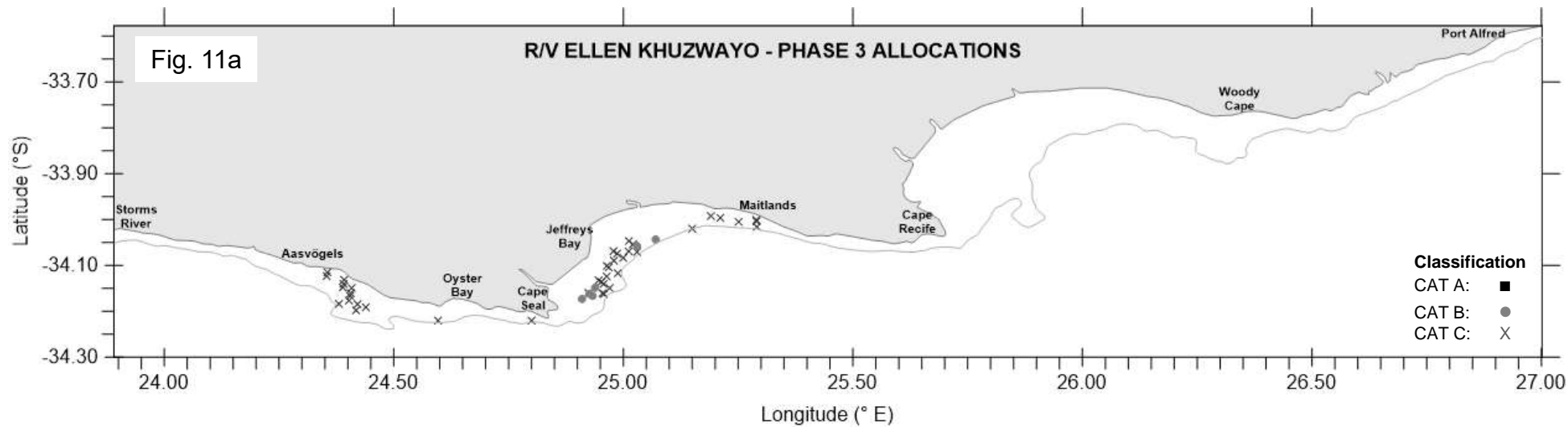


Fig. 11: Location of the A, B and C- Category marks in Phase 3 (Fig. 11a) and Phase 4 (Fig. 11b) of the *Ellen Khuzwayo* survey.

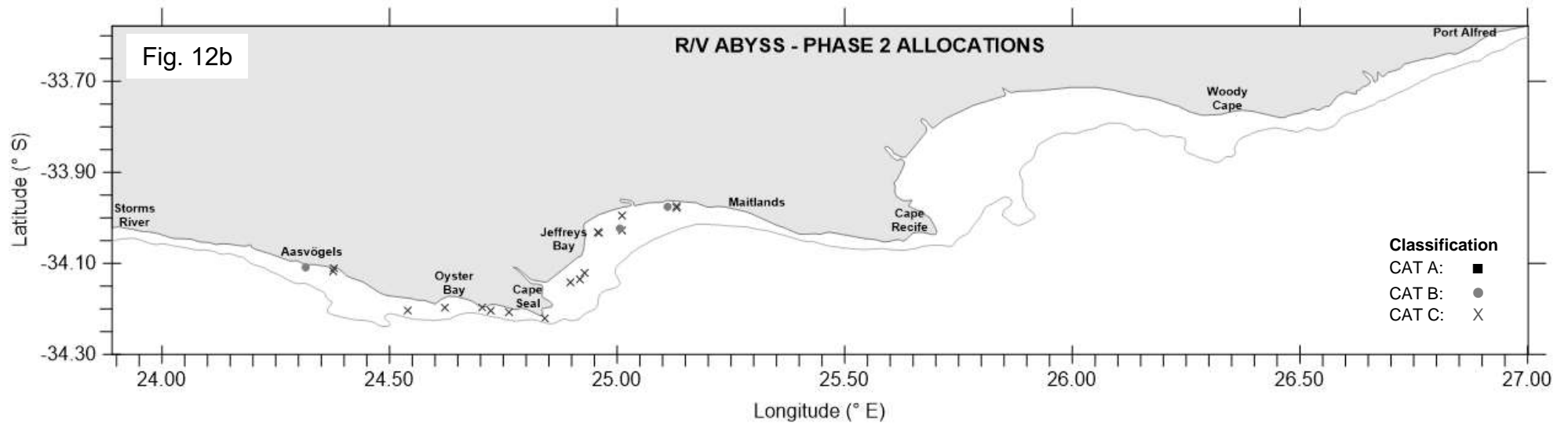
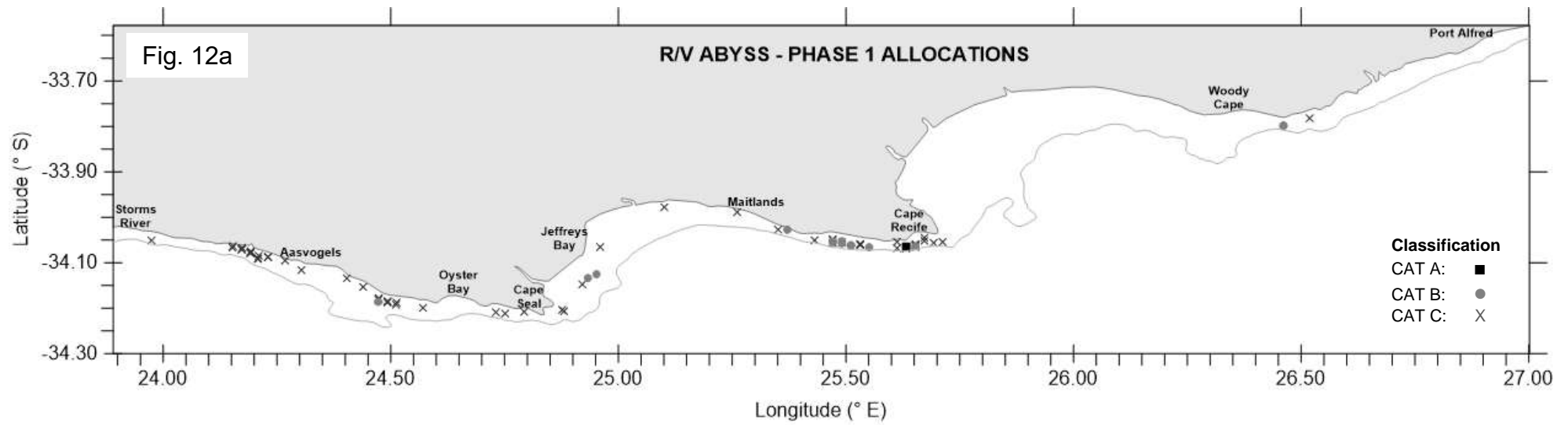


Fig. 12: Location of the A, B and C- Category marks in Phase 1 (Fig. 12a) and Phase 2 (Fig. 12b) of the *Abyss* survey.

| R/V ELLEN KHUZWAYO: Squid Acoustic Biomass Surveys (Depth 30 – 50m) – 06 to 20 November 2021. | | | | | | | |
|--|---------------------------|------------------------|---------------------------|---------------------------|--------------------------|---------------------------|---------------------------|
| STRATUM | 1A | 1B | 2A | 2B | 2C | 3 | 4 |
| AREA | Storms River to Aasvögels | Aasvögels to Cape Seal | Cape Seal to Jeffreys Bay | Jeffreys Bay to Maitlands | Maitlands to Cape Recife | Cape Recife to Woody Cape | Woody Cape to Port Alfred |
| PHASE 1 – TONNES (CV) | | | | | | | |
| Date | | 06/11 | 07/11 | 07/11 | | 08/11 | 10/11 |
| Cat A | | 0 | 0 | 0 | | 0 | 0 |
| Cat B | | 84.4 (0.57) | 0 | 92.3 (0.73) | | 63.6 (0.51) | 43.4 (1.01) |
| Cat C | | 124.1 (0.93) | 103.0 (0.97) | 95.9 (0.45) | | 170.6 (0.49) | 397.4 (0.44) |
| PHASE 2 – TONNES (CV) | | | | | | | |
| Date | 11/11 | 12/11 | 13/11 | 13/11 | 14/11 | 14-16/11 | 16/11 |
| Cat A | 0 | 18.3 (1.02) | 190.6 (0.99) | 135.2 (0.63) | 0 | 0 | 0 |
| Cat B | 0 | 2.90 (1.04) | 206.4 (0.46) | 160.4 (0.35) | 0 | 68.5 (0.56) | 30.4 (0.58) |
| Cat C | 93.3 (0.72) | 215.6 (0.35) | 86.0 (0.55) | 55.6 (0.51) | 69.0 (0.43) | 215.1 (0.38) | 65.8 (0.44) |
| PHASE 3 – TONNES (CV) | | | | | | | |
| Date | | 19/11 | 17/11 | 17/11 | | | |
| Cat A | | 0 | 0 | 0 | | | |
| Cat B | | 0 | 55.5 (0.63) | 48.6 (0.66) | | | |
| Cat C | | 235.9 (0.40) | 41.1 (0.84) | 109.6 (0.53) | | | |
| PHASE 4 – TONNES (CV) | | | | | | | |
| Date | | 20/11 | 18/11 | 18/11 | | | |
| Cat A | | 0 | 228.1 (0.99) | 353.8 (1.02) | | | |
| Cat B | | 0 | 166.9 (0.59) | 61.7 (0.64) | | | |
| Cat C | | 118.6 (0.31) | 88.7 (0.72) | 165.5 (0.44) | | | |

Table 5: Estimates of squid biomass (in tonnes) and CVs (parenthesis) made from the A, B and C - Category marks for all strata in the *Ellen Khuzwayo* survey. See Table 2 for the dates and geographical limits of each survey. As in Table 2, surveys of strata with the same number are nominally replicates.

| R/V ABYSS: Squid Acoustic Biomass Surveys (Depth 15 – 30m) – 31 October to 15 November 2021. | | | | | | | |
|---|---------------------------|------------------------|---------------------------|---------------------------|--------------------------|---------------------------|---------------------------|
| STRATUM | 1A | 1B | 2A | 2B | 2C | 3 | 4 |
| AREA | Storms River to Aasvögels | Aasvögels to Cape Seal | Cape Seal to Jeffreys Bay | Jeffreys Bay to Maitlands | Maitlands to Cape Recife | Cape Recife to Woody Cape | Woody Cape to Port Alfred |
| PHASE 1 – TONNES (CV) | | | | | | | |
| Date | 31/10 | 03/11 | 04/11 | 06/11 | 07/11 | | 10/11 |
| Cat A | 0 | 0 | 0 | 0 | 112.1 (1.01) | | 0 |
| Cat B | 0 | 39.3 (1.00) | 131.0 (0.67) | 0 | 151.3 (0.33) | | 5.7 (0.99) |
| Cat C | 146.5 (0.33) | 160.7 (0.63) | 91.1 (0.67) | 52.0 (0.79) | 189.5 (0.60) | | 10.5 (0.98) |
| PHASE 2 – TONNES (CV) | | | | | | | |
| Date | | 12/11 | 14/11 | 15/11 | | | |
| Cat A | | 0 | 0 | 0 | | | |
| Cat B | | 3.2 (1.01) | 10.6 (0.96) | 23.2 (1.00) | | | |
| Cat C | | 73.6 (0.40) | 35.3 (0.46) | 41.8 (0.65) | | | |

Table 6: Estimates of squid biomass (in tonnes) and CVs (parenthesis) made from the A, B and C - Category marks for all strata surveyed by *Abyss*. See Table 3 for the dates and geographical limits of each survey. As in Table 3, surveys of strata with the same number are nominally replicates.

In Table 7, the biomass estimates made from A, B and C - Category marks are summarised for both vessels, for each phase of the survey; in each case by summing the biomass estimates for the individual strata in Tables 5 and 6. Also shown (in parenthesis) are the associated CVs, obtained from the CVs in Tables 5 and 6. The final columns show the sum of the estimates for both vessels for Phases 1 and 2, these being the surveys which were surveyed at more or less the same time by both vessels.

| Category | <i>Ellen Khuzwayo</i> | | | | <i>Abyss</i> | | <i>Ellen Khuzwayo + Abyss</i> | |
|----------|-----------------------|-------------------|-----------------|-------------------|-------------------|-----------------|-------------------------------|------------------|
| | Phase 1 | Phase 2 | Phase 3 | Phase 4 | Phase 1 | Phase 2 | Phase 1 | Phase 2 |
| A | 0 | 344.1 (0.60) | 0 | 581.9 (0.73) | 112.1 (1.01) | 0 | 112.1 (1.01) | 344.1 (0.60) |
| B | 283.7 (0.35) | 468.6 (0.25) | 104.1 (0.46) | 228.6 (0.46) | 327.3 (0.33) | 37 (0.69) | 611.0 (0.24) | 505.6 (0.24) |
| C | 891.0 (0.20) | 800.4 (0.18) | 386.6 (0.30) | 372.8 (0.28) | 650.3 (0.26) | 150.7 (0.29) | 1 541.3 (0.16) | 951.1 (0.18) |
| Total | 1 174.7 (0.15) | 1 613.1 (0.16) | 490.7 (0.34) | 1 183.3 (0.38) | 1 089.7 (0.21) | 187.7 (0.27) | 2 264.4 (0.14) | 1665.8 (0.17) |

Table 7: Summary of biomass estimates (in tonnes) and CVs (parenthesis) in each of the three identification categories, for all phases of the survey, for both vessels, plus the sum of the estimates for Phases 1 and 2.

Table 8 gives the estimates and CVs for the core strata, which were surveyed by both vessels in Phase 1 and Phase 2 - ie. Strata 1B, 2A and 2B (Aasvögels Bult to Maitlands), and Table 9 the totals and CVs in all three identification categories for both vessels for these strata.

| Stratum | Category | Phase 1 | | Phase 2 | |
|---------|----------|-----------------------|--------------|-----------------------|--------------|
| | | <i>Ellen Khuzwayo</i> | <i>Abyss</i> | <i>Ellen Khuzwayo</i> | <i>Abyss</i> |
| | | Biomass (CV) | Biomass (CV) | Biomass (CV) | Biomass (CV) |
| 1B | A | 0 | 0 | 18.30 (1.02) | 0 |
| | B | 84.4 (0.56) | 39.3 (1.00) | 2.90 (1.04) | 3.2 (1.00) |
| | C | 124.1 (0.93) | 160.7 (0.65) | 215.6 (0.35) | 73.6 (0.40) |
| | All | 208.5 (0.60) | 200.0 (0.55) | 236.9 (0.33) | 76.80 (0.39) |
| 2A | A | 0 | 0 | 190.6 (0.99) | 0 |
| | B | 0 | 131.0 (0.67) | 206.4 (0.45) | 10.60 (0.96) |
| | C | 103.0 (0.97) | 91.1 (0.67) | 86.0 (0.55) | 35.30 (0.45) |
| | All | 103.0 (0.97) | 222.1 (0.48) | 483 (0.45) | 45.90 (0.41) |
| 2B | A | 0 | 0 | 135.2 (0.63) | 0 |
| | B | 92.3 (0.73) | 0 | 160.4 (0.35) | 23.20 (1.00) |
| | C | 95.9 (0.45) | 52.0 (0.79) | 55.6 (0.514) | 41.80 (0.65) |
| | All | 440.8 (0.40) | 52.0 (0.79) | 351.2 (0.30) | 65.00 (0.55) |
| Totals | | 752.3 (0.32) | 474.1 (0.33) | 1071.1 (0.24) | 187 (0.27) |

Table 8: Biomass estimates (in tonnes) and CVs (parenthesis) for strata surveyed by both *Ellen Khuzwayo* and *Abyss* in Phases 1 and 2, in all three identification categories.

| Category | <i>Ellen Khuzwayo</i> | | | | <i>Abyss</i> | |
|----------------|-----------------------|-------------------|-----------------|-------------------|-----------------|-----------------|
| | Phase 1 | Phase 2 | Phase 3 | Phase 4 | Phase 1 | Phase 2 |
| A | 0 | 334.1 (0.60) | 0 | 581.9 (0.73) | 0 | 0 |
| B | 176.7 (0.47) | 369.7 (0.30) | 165.1 (0.41) | 228.6 (0.46) | 170.3 (0.56) | 37.0 (0.69) |
| C | 319.4 (0.52) | 357.2 (0.26) | 386.6 (0.30) | 372.8 (0.27) | 303.8 (0.41) | 150.7 (0.29) |
| All Categories | 495.7 (0.37) | 1 061.0 (0.23) | 551.7 (0.24) | 1 183.4 (0.38) | 474.1 (0.33) | 187.7 (0.27) |

Table 9: Biomass and CV estimates in each identification category in the core strata which were surveyed by *Ellen Khuzwayo* and *Abyss*.

For each vessel, the estimates for the different Phases in Table 9, which are nominally replicates, were averaged and added to the single estimates for the other strata to give estimates of total biomass in the three identification categories for that vessel. These were then added to give the estimates of total biomass for the whole survey. The resulting biomass estimates and corresponding CVs are set out in Table 10.

| Category | <i>Ellen Khuzwayo</i> | | <i>Abyss</i> | | <i>Ellen Khuzwayo + Abyss</i> | |
|----------------|-----------------------|------|------------------|------|-------------------------------|------|
| | Biomass (tonnes) | CV | Biomass (tonnes) | CV | Biomass (tonnes) | CV |
| A | 231.5 | 0.51 | 112 | 1.0 | 343.5 | 0.47 |
| B | 322.7 | 0.17 | 260.6 | 0.27 | 583.3 | 0.15 |
| C | 864.6 | 0.15 | 573.7 | 0.25 | 1 438.3 | 0.13 |
| All Categories | 1 419.1 | 0.13 | 946.4 | 0.20 | 2 022.0 | 0.09 |

Table 10: Biomass estimates in all three identification categories and CVs for both vessels. Mean estimates for strata surveyed more than once have been averaged and added to the estimates for those strata surveyed only once.

Finally, Table 11 gives the same information for various combinations of the identification categories, including a combination of Category A and half Category B, as in the analysis of the results from the 2020 survey (Soule and Hampton, 2021). The estimates are for all strata, including those which were surveyed by only one of the vessels, and for the core strata, which were surveyed by both vessels.

| Category | <i>Ellen Khuzwayo</i> | | <i>Abyss</i> | | <i>Ellen Khuzwayo + Abyss</i> | |
|----------|-----------------------|------|------------------|------|-------------------------------|------|
| | Biomass (tonnes) | CV | Biomass (tonnes) | CV | Biomass (tonnes) | CV |
| A | 231.5 | 0.51 | 112 | 1.0 | 343.5 | 0.47 |
| B | 322.7 | 0.17 | 260.6 | 0.27 | 583.3 | 0.15 |
| C | 864.6 | 0.15 | 573.7 | 0.25 | 1438.3 | 0.13 |
| A+B | 554.3 | 0.23 | 260.6 | 0.27 | 926.8 | 0.20 |
| A+B/2 | 392.8 | 0.33 | 130.3 | 0.18 | 635.1 | 0.20 |
| A+B+C | 1 419.1 | 0.13 | 946.4 | 0.20 | 2 022.0 | 0.09 |

Table 11: Biomass and CV estimates for the entire survey derived from different combinations of the A, B and C estimates in Table 10.

Discussion

This survey was by far the most comprehensive acoustic survey of *L. reynaudii* yet undertaken, both in terms of duration (14 days for *Ellen Khuzwayo* and 16 days for *Abyss*) and the stretch of coastline covered (Storms River to Port Alfred). This is effectively the full extent of the inshore commercial fishing grounds – the first time these grounds have been comprehensively surveyed acoustically. This is in contrast to the survey by *Ellen Khuzwayo* in 2019, which surveyed between Aasvögels Bult and Cape Recife over a period of 9 days, and the survey by *Abyss* in 2020, which covered the area between Cape Seal and Maitlands within the 50 m isobath (to a minimum depth of about 15m) sporadically over a period of 18 days. The extension of the survey area to as far west as the Storms River mouth and eastwards to Port Alfred, has added new information on these hitherto un-surveyed areas. Note too, that this is the first time that the inshore zone within the 30 m isobath has been comprehensively and systematically surveyed over such a large section of coastline in support of a survey over the same length of coastline further offshore at much the same time. The long duration of the two surveys enabled the core area (Aasvögels Bult to Maitlands) to be surveyed more than once by both vessels (four times in the case of *Ellen Khuzwayo* and twice in the case of *Abyss*) to gather information about the dynamics of the population in this key fishing area over a period of two weeks.

The A marks were confidently identified, but were few in number (viz; only 11 in the entire survey). With the exception of one mark on the periphery off Cape Recife (see Fig 12a) they fell within the area of highest commercial catches at this time of the year. Nonetheless, they contributed 37% of the estimate of total biomass in the A and B categories and 17% of the estimate of total biomass in all three categories (Table 11). The extreme patchiness of their distribution is reflected in the high CVs of the A - Category estimates (see Tables 5 and 6, for example), which approached, and even exceeded, 100% in places, and in the estimate of total A - Category biomass in Table 11 (47%). It is also evident in the large variation in biomass and the high CVs for A - Category marks in the core strata (Table 9).

Figs. 10 to 12 and, for example, Table 7, show that the B - Category marks, which were less confidently identified, but which were nonetheless believed to be probably squid, were much more widely spread than the A - Category marks. This is reflected in the CV estimates, which were typically about half of those of the A - Category estimates. There were 71 of them in total, most of which were concentrated within the main commercial fishing ground, particularly between Cape Seal and Maitlands (Figs. 10 to 12). At times they were in the vicinity of the A - Category marks, but this may be partly an artefact of the fact that one of the criteria for classifying a mark as B was whether or not there was an A - Category mark nearby (see, for example, Fig. 6).

In contrast to the A - Category, and to a less extent, the B - Category marks, the C - Category marks were very widely spread throughout the survey area (see Figs. 10 to 12) and, for example, Table 7. They were far more numerous, with 231 in total having been tentatively identified. Tables 11 to 13 shows that, as would be expected, the CVs in these estimates were generally substantially lower than those of the A - and B - Category estimates. The widespread nature of the marks and their lack of any clear association with the A - or B - Category marks, particularly to the west of Cape Seal (cf. Figs. 10 and 11.) suggests that many of them were probably not squid.

Of the various combinations of the A, B and C estimates in Table 11, we regard the A estimate (343.5 tonnes) as a minimum estimate, since it is unlikely that all spawners would be in actively spawning aggregations at any one time. Conversely, since it is certain that many (perhaps most) of the C - Category marks were not squid, the estimate for all three categories (2 022 tonnes) should be seen as an extreme upper limit. Since at least some of the B - Category marks (which we regarded as *probably* squid) would have been squid, they need to be taken into account to some extent. We have chosen, as did Soule *et al.* (2021), to include half of their estimated biomass in the final survey estimate, which minimises the maximum error from including them. We therefore propose this estimate (635 tonnes, with a CV of 20 % - Table 11) as the most reasonable estimate of spawning squid biomass for the survey. The CV is acceptably low for an acoustic survey, despite the large CVs in most of the A and B estimates, largely because of the large number of data points on which they are based - a consequence of the exceptionally large amount of survey effort expended by both vessels. This estimate should however be treated with caution as an estimate of precision because of other uncertainties - particularly that in species identification.

In Table 12 the current estimates for Strata 2A and 2B combined (Cape Seal to Maitlands) are compared with the estimates for roughly the same area from the 2019 and 2020 surveys. Note that the 2020 estimates by *Abyss* include the inshore strata surveyed by *Abyss* in the current survey, since the inshore region was in effect covered in the 2020 survey. Note also that no attempt was made to include marks other than A - Category marks in 2019, nor to include C - Category marks in either 2019 or 2020.

| Year | Vessel | Biomass A (tonnes) | Biomass B (tonnes) | Biomass (A + B/2) (tonnes) |
|------|-----------------------|--------------------|--------------------|----------------------------|
| 2019 | <i>Ellen Khuzwayo</i> | 49.3 (0.41) | - | - |
| 2020 | <i>Abyss</i> | 221.5 (0.27) | 140.8 (0.19) | 291.8 (0.21) |
| 2021 | <i>Ellen Khuzwayo</i> | 226.8 (0.51) | 291.8 (0.21) | 372.7 (0.25) |

Table 12: Comparison between various biomass estimates and CVs for Strata 2A and 2B combined in the 2019, 2020 and 2021 surveys. The estimates from the 2019 and 2020 surveys have been calculated from data in Soule *et al.* (2020) and Soule *et al.*, (2021) respectively. All 2021 estimates have been calculated from those in Table 5 and 6.

Table 12 shows that while the (A+B/2) estimate and associated CV from the current survey is similar to the estimate and CV from the 2020 survey, there is a 4.6 – fold difference in the A - Category estimates between the current survey and that in 2019, although the CVs are roughly similar. Some of this difference could be due to the low precision of both estimates (as reflected in the high CVs), but we note that in 2019, 94% of the estimated A - Category biomass came from Stratum 1B, west of Cape Seal (Aasvögels Bult to Cape Seal), which was not surveyed by *Abyss* in 2020, which suggests a major difference in distribution at the time of the 2019 and 2021 surveys. Overall, therefore, we consider that there is little evidence from this limited subset of the data to indicate whether the biomass in the current survey was greater or less than in the two previous years. Nonetheless, the similarity in the biomass estimates in the core area in 2020 and 2021, and in the nature of the marks in all three years (see for example, marks in Soule and Hampton, 2020 and 2021), does suggest that the broad behaviour of the squid was similar in the three years, particularly in the core area where A – and B - Category densities were highest, and where catches are commonly higher than elsewhere (Fig. 13).

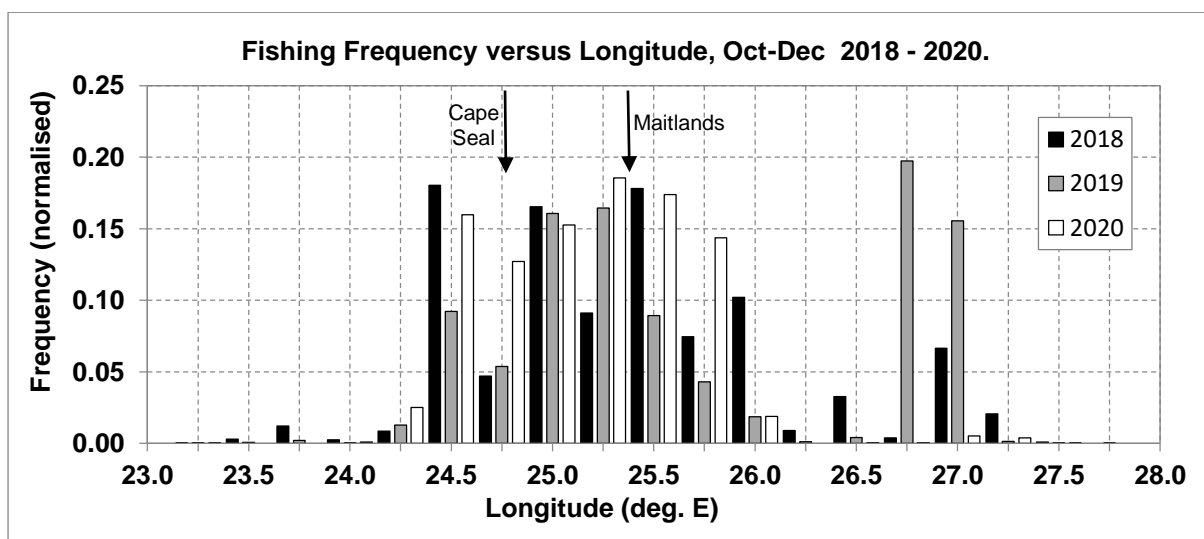


Fig. 13: Fishing effort in October, November and December between 2018 and 2020 in relation to longitude. Data courtesy Dr Jean Waruguru Mwiggi, DEFF.

Comparison between the Phase 1 and Phase 2 estimates in Table 9 for the *Ellen Khuzwayo* survey, and between Phases 1 and 2 for the survey by *Abyss*, shows that while the former increased between Phases 1 and 2, with no A marks whatsoever in Phase 1, the opposite was true for the *Abyss* survey. This may indicate that there was a general movement into deeper water between Phases 1 and 2 of the survey (which were roughly a week apart for both vessels), although the evidence is not particularly strong. Similarly the fact that the estimates in Table 9 from Phase 4 of the *Ellen Khuzwayo* survey are on average roughly double those from Phase 3, some 3 days earlier (Table 2), with again, no A - Category marks detected in the earlier phase, could again be evidence (again albeit weak) of a movement into deeper water between the two phases. Overall however, there is no strong evidence of a large-scale immigration to, or emigration, from the inshore spawning grounds during the two-week period of the survey.

The fact that the 38 - kHz target strength per kg estimates in Table 4 are close to the means in the long-term study by Soule *et al.* (2010) over a long period, is evidence of the consistency of *L. reynaudii* aggregations as acoustic targets over a long time period. This increases the credibility and value of the surveys as a monitoring tool, at least if restricted to the A and, probably, the B aggregations.

It is of interest to compare the 2021 estimates with the commercial jig catches in 2021; particularly those made relatively soon before and after the surveys. These catches are shown by month in Table 13.

| Jan | Feb | March | April | May | June | July | Aug | Sept | Oct | Nov | Dec |
|-------|-------|-------|--------------------------|-----|------|-------|------|-------|-------|------|-------|
| 930.1 | 323.3 | 303.9 | Additional Closed Season | | | 289.7 | 84.7 | 506.4 | 509.8 | 18.9 | 379.4 |

Table 13: Commercial jig catches (in tonnes) by month in 2021. Note that there were only 9 fishing days in November because of the closed season in that month. Data courtesy Dr Jean Waruguru Mwicigi, DEFF.

It is particularly noticeable that the monthly catch in the nine fishing days in November (18.9 tonnes) was by far the lowest in the year, suggesting an unusually low abundance of spawning aggregations then, even given the fact that weather conditions were apparently poor for much of this time. We note further that the total catch in 2021 (3 346 tonnes) was the second lowest since 2007; second only to the anomalously low catch of 2 647 tonnes in 2013. This catch, and the catches of 510 tonnes and 379 tonnes in October and December respectively, are consistent with the A+B/2 estimate in Table 11 for the two vessels combined (635.1 tonnes, with a CV of 20 %), which does add some additional credence to the estimate. Certainly, there is nothing in the catches *per se* to cast doubt on the estimate.

We believe that the current survey estimate is credible as an estimate of the biomass of aggregated squid on the traditional inshore fishing grounds at the time of the survey. However, little can be said at this stage about how this relates to the size of the adult population at large, since the proportion of the population which is elsewhere at the time and/or which is perhaps acoustically undetectable, is unknown. This is crucial to the broader

question of whether, and if so, how, acoustic estimates of biomass in November can be used in management of the resource, either as relative indices or as absolute estimates. This question is one which will require greater understanding of the large-scale movement and aggregating behaviour of the population at other times of the year to supplement what has already been learned from the more limited studies to date. Answering it is clearly beyond the scope of the current study, but we suggest that one approach might be to examine seasonal changes in diurnal catch rates from the large body of existing commercial data, which could provide valuable information on changes in the aggregated proportion throughout the year, at least.

It must be appreciated that the acoustic survey programme *per se* is still in its early stages, with the current survey being the first full-scale survey of the inshore commercial grounds, built on the more limited pilot surveys in the two previous years. Continuation of the programme, and its further development as a monitoring tool, will require standardisation on the current survey design, and particularly further methodological studies on the crucial question of target identification by whatever means possible.

Conclusions

Our major conclusions from the survey can be summarised as follows:

1. The survey was highly successful in that it demonstrated that the entire inshore squid fishing ground between about 15 m and 50 m can be surveyed acoustically on lines as little as 0.5 n.mile apart within the November closed season, with existing equipment, vessels and personnel.
2. While the extension of the survey area to the west of Aasvögels Bult and to the east of Cape Recife did reveal the presence of squid in these areas, the proportion of the biomass there was low compared to that in the core area where most fishing activity takes place, and where the previous two surveys were concentrated. This suggests that future acoustic surveys could profitably be restricted to the area between Aasvögels and Cape Recife, to increase efficiency.
3. In contrast, the survey, for the first time, of the entire inshore region to a minimum depth of 10 m by *Abyss* showed that there can be significant quantities of squid in water shallower than 30 m. Clearly, it will be important to continue surveying this area with a small vessel in future surveys.
4. What we consider to be our best estimate of the squid biomass within the 50 m isobath between Storms River mouth and Port Alfred, including the near shore zone (635 tonnes with a CV of 20%) is consistent with catches shortly after the survey and with the A and B estimates from the survey of a smaller area from *Ellen Khuzwayo* in 2019 and *Abyss* in 2020, using the same methods. Both support the estimates in this study to some extent.
5. The addition of the C- Category marks for the first time, as an exercise to examine the potential error due to a significant proportion of the squid biomass being in small, scattered schools unlike those in characteristic spawning aggregations raises the

estimate to 2 022 tonnes and reduces the CV to 9 %. However, due to the uncertainty in classifying these marks in the presence of many similar marks from pelagic fish and other species, and the lack of direct evidence at this stage that *L. reynaudii* can indeed form these kinds of mark in abundance on the inshore spawning grounds, we tentatively conclude that this estimate is positively biased, perhaps by a large amount. As such it is probably safe to regard it as an absolute upper limit, for whatever purpose this may serve at present. Whether it will be worthwhile to include the C marks in any future analysis, given that it greatly increases the complexity of the analysis and, because of the large number of marks which have to be scrutinised by multiple criteria, the time needed to complete the report, is debateable.

6. Differences in the biomass estimates between the various phases may be partly due to movement of the population within the survey area, but the evidence for this is weak, largely due to the large CVs in the Stratum estimates. Overall, there is nothing to suggest a large-scale immigration or emigration of the spawning population to or from the survey area during the two-week period of the survey.
7. The question of whether the current estimate is a reliable absolute or relative estimate of the size of the entire adult population on the inshore spawning ground, and therefore of its potential use for management at this stage, is beyond the scope or brief of this study. We believe that such a broad question would best be addressed, *inter alia*, through an in-depth examination of the large amount of data available from the commercial fleet on the changes in the position and diurnal catch patterns throughout the year. It should also be profitable to continue attempting to compare the estimates with the commercial catches immediately before and after the survey as a form of ground-truthing.
8. The design and survey methods used in this survey should become the standard for future acoustic surveys of *L. reynaudii* during the November closed season to optimise comparison of the estimates between the years. In any such surveys, it will be important to continue attempting to improve confidence in target recognition, particularly for the weaker targets.

Acknowledgements

We gratefully acknowledge the co-operation and professionalism of Captain Raul Mulligan and crew of *R. V. Ellen Khuzwayo* in maintaining accurate courses and accommodating all navigational requests throughout what was a lengthy and exacting survey. We are equally grateful to Johan Rademan and his team on Research Inflatable *Abyss* for the excellent work they put into preparing the vessel, and the extremely long survey hours they worked, often under difficult circumstances, which has enabled us to obtain an estimate of squid biomass within the near-shore zone for the first time. Finally, we thank DFFE (courtesy Dr. Jean Waruguru Mwicigi) for the catch information in Fig. 13 and Table 13.

References

- Ona, E. and Mitson, R. B. 1996. Acoustic sampling and signal processing near the seabed: the deadzone revisited. *ICES Journal of Marine Science*, 53: 677–690.
- Jolly, G.M. and I. Hampton 1990. A stratified random transect design for acoustic surveys of fish stocks. *Canadian Journal of Fisheries and Aquatic Sciences* 47(7): 1282- 1291.
- Lipinski, M. R. 1994. Differences among basic biological parameters in a population of chokka squid (*Loligo vulgaris reynaudii*, Cephalopoda: Loliginidae) sampled by three methods. *South African Journal of Marine Science*, 14: 281-286.
- Simmonds, J. and D.N. MacLennan 2006- *Fisheries Acoustics: Theory and Practice* (2nd Edition). ISDN: 978-632-05994-2. 456 pp.
- Soule, M. A., Hampton, I., and Lipinski, M. R. 2010- Estimating the target strength of live, free-swimming chokka squid *Loligo reynaudii* at 38 and 120 kHz. *ICES Journal of Marine Science*, 67: 1381–1391.
- Soule, M. A. and I. Hampton 2020. Acoustic survey of chokka squid (*Loligo reynaudii*) from *R.V. Ellen Khuzwayo* 30 October to 9 November 2019. Report by Fisheries Resource Surveys cc. Cape Town to SASMIA, 36 pp.
- Soule, M. A. and I. Hampton 2021. Acoustic survey of chokka squid on inshore spawning grounds between Seal Point and Maitlands in the November 2020 closed season from Research Inflatable *Abyss*. Report by Fisheries Resource Surveys cc. Cape Town to SASMIA, 10 pp.

Appendix A

Acoustic Calibration Report: *R.V. Ellen Khuzwayo*

| R/V <i>Ellen Khuzwayo</i> Calibration – Plettenberg Bay - 05 November 2021. | | | | |
|---|------------------------------|--------------------|-----------------------------|--------------------|
| Frequency | 38 kHz | | 200 kHz | |
| Transducer Model | ES38-B | | ES200-7C | |
| 38.1 mm WC Sphere TS (dB) | -42.3 | | -38.9 | |
| Power (watts) | 2000 | | 150 | |
| Pulse Duration (ms) | 0.512 | | 0.512 | |
| Absorption (dB/km) | 8.5 | | 67.8 | |
| Sound Speed (m/s) | 1513 | | 1513 | |
| Transceiver Parameters | EK60 38kHz GPT Initial | LOBE Calibrated | EK60 200 kHz GPT Initial | LOBE Calibrated |
| G ₀ (dB) | 26.00 | 25.17 | 27.00 | 25.24 |
| S _A Correction (dB) | 0.00 | -0.62 | 0.00 | -0.34 |
| S _V Gain (dB) | 26.00 | 24.55 | 27.00 | 24.90 |
| Alongships 3dB Beam Width | 6.97 ° | 6.99 ° | 6.35 ° | 6.43 ° |
| Alongships Offset | -0.01 ° | -0.0 ° | +0.05 ° | -0.10 ° |
| Ahwardships 3dB Beam Width | 6.92 ° | 6.98 ° | 6.39 ° | 6.39 ° |
| Athwardships Offset | -0.05 ° | +0.01 ° | +0.03 ° | +0.16 ° |
| RMS Error: Beam Model (dB) | — | 0.17 | — | 0.26 |
| RMS Error: Poly. Model (dB) | — | 0.15 | — | 0.22 |

Table A-1: Results obtained at 38 and 200 kHz for the calibration carried out off Robberg, Plettenberg Bay, on *R/V Ellen Khuzwayo* immediately prior to the start of the survey on 05 Nov. 2021.

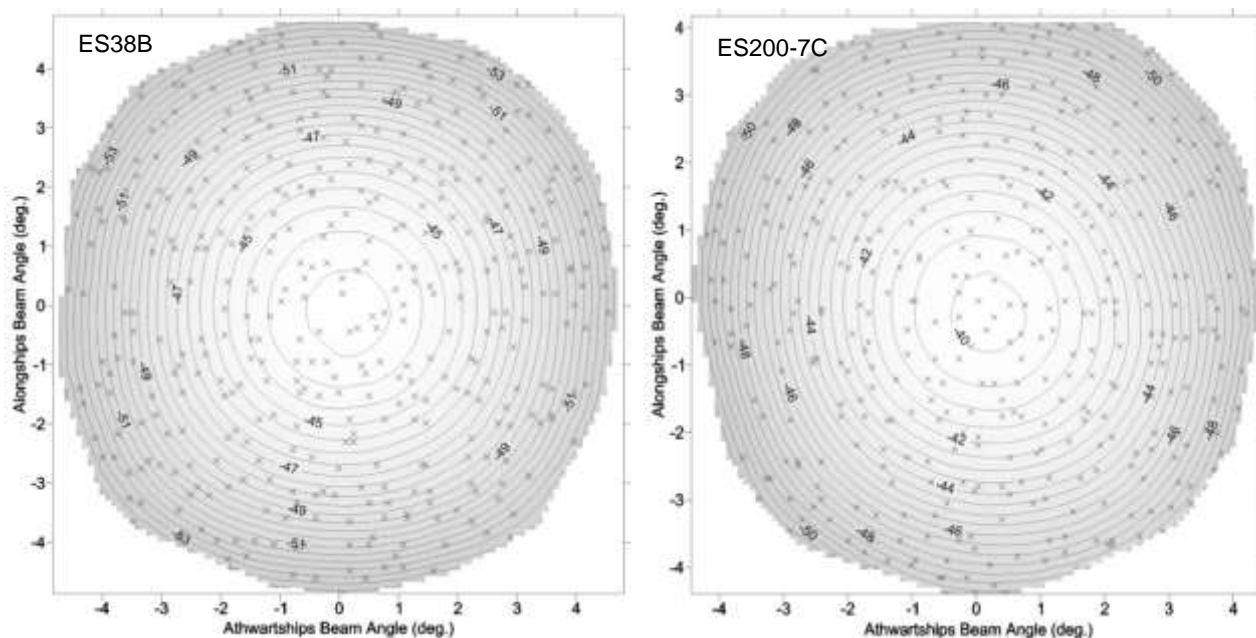


Fig. A-1: Contoured beamplots for the ES38B and ES200-7C transducers derived from data logged by the SIMRAD Lobe calibration routine during the calibration exercise. Contours are separated by 0.5 dB. Crosses mark the position of sphere detections recorded across the beams.

Appendix B

Acoustic Calibration Report: Research Inflatable *Abyss*

| R/V <i>Abyss</i> Calibrations | | | | |
|--------------------------------|---------------------------|--------------------|----------------------------|--------------------|
| Date | 16 April 2021 | | 17 May 2022 | |
| Location | Oudekraal – Cape Town | | Kromme – St. Francis Bay | |
| Frequency | 38 kHz | | | |
| Transducer Model | ES38-B | | ES38-B | |
| 38.1 mm WC Sphere TS (dB) | -42.3 | | -42.3 | |
| Power (watts) | 1000 | | 1000 | |
| Pulse Duration (ms) | 0.512 | | 0.512 | |
| Absorption (dB/km) | 8.9 | | 7.7 | |
| Sound Speed (m/s) | 1509 | | 1521 | |
| Transceiver Parameters | EK60 38kHz GPT Initial | LOBE Calibrated | EK60 38 kHz GPT Initial | LOBE Calibrated |
| G ₀ (dB) | 20.91 | 20.63 | 20.59 | 20.61 |
| S _A Correction (dB) | -0.80 | -0.65 | -0.75 | -0.76 |
| S _V Gain (dB) | 20.11 | 19.98 | 19.84 | 19.85 |
| Alongships 3dB Beam Width | 12.11° | 12.13° | 12.11 ° | 11.97 ° |
| Alongships Offset | -0.07° | -0.13° | -0.07 ° | -0.06 ° |
| Ahwardships 3dB Beam Width | 12.15° | 12.07° | 12.15 ° | 11.90 ° |
| Athwardships Offset | +0.09° | -0.02° | +0.09 ° | +0.02 ° |
| RMS Error: Beam Model (dB) | — | 0.26 | — | 0.21 |
| RMS Error: Poly. Model (dB) | — | 0.23 | — | 0.19 |

Table B-1: Results obtained at 38 kHz for calibrations carried out on R/V *Abyss* on 16 April 2021, off Oudekraal, Cape Town, and on 17 May 2022, in St Francis Bay.

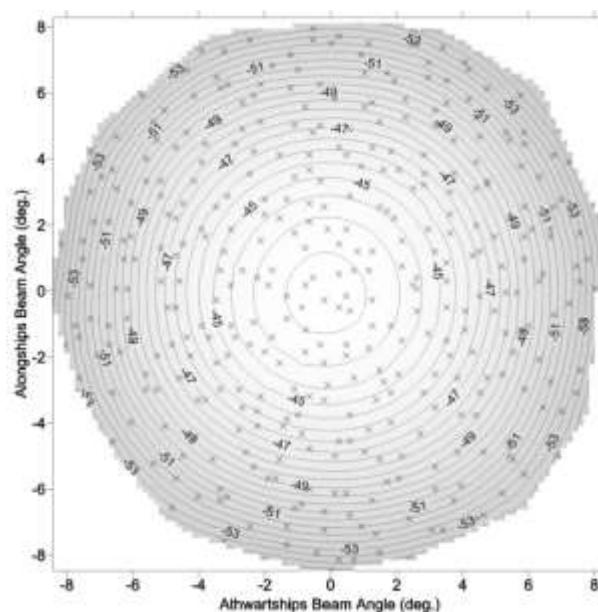


Fig. B-1: Contoured beamplot for the ES38-12 transducer deployed on R/V *Abyss* derived from data logged by the SIMRAD Lobe routine during the calibration exercise on 17 May 2022. Contours are separated by 0.5 dB. Crosses mark the position of sphere detections recorded across the beam.

Appendix C

Biological data collected from *F.V. Silver Eagle* and *F.V. Silver Laguna*

| Vessel | Date (2021) | Time Start/End | Position Lat.°S/ Long.°E | Sample Weight (g) | N _m / N _f | N _{TOT} | W (g) | ML (cm) |
|--------------------------|-------------|---------------------------------|----------------------------------|-------------------|---------------------------------|------------------|-------|---------|
| F/V SILVER EAGLE | 03/11 | 14:55 18:41 | 34 ° 02.1571 ' 25 ° 03.8101 ' | 27500 | 98 / 17 | 115 | 201 | 22.6 |
| | 04/11 | 06:40 09:03 | 34 ° 02.1224 ' 25 ° 03.7973 ' | 32000 | 90 / 17 | 107 | 237 | 22.8 |
| | 04/11 | 10:15 16:50 | 34 ° 03.8949 ' 25 ° 01.1324 ' | 32000 | 93 / 11 | 104 | 252 | 23.4 |
| | 04/11 | 13:14 16:50 | 34 ° 04.4099 ' 24 ° 58.6832 ' | 34500 | 101 / 2 | 103 | 293 | 23.5 |
| | 05/11 | 08:32 12:06 | 34 ° 04.4451 ' 24 ° 58.6769 ' | 30500 | 95 / 5 | 100 | 255 | 21.7 |
| | 06/11 | 09:45 11:18 | 34 ° 09.3522 ' 24 ° 52.5348 ' | 30500 | 103 / 12 | 115 | 240 | 21.3 |
| | 06/11 | 15:13 17:00 | 34 ° 04.4230 ' 24 ° 58.6769 ' | 32000 | 103 / 2 | 105 | 278 | 21.6 |
| | 08/11 | 06:10 07:40 | 34 ° 09.3454 ' 24 ° 52.5276 ' | 32000 | 89 / 11 | 100 | 279 | 22.6 |
| | 08/11 | 17:30 17:47 | 34 ° 03.4804 ' 25 ° 40.3825 ' | 33800 | 111 / 1 | 112 | 301 | 22.3 |
| | 09/11 | 17:23 20:53 | 33 ° 59.8690 ' 25 ° 14.5467 ' | 33000 | 94 / 11 | 105 | 256 | 22.5 |
| | 10/11 | 06:25 06:49 | 33 ° 59.5427 ' 25 ° 13.3649 ' | 31500 | 99 / 2 | 101 | 282 | 22.6 |
| | 10/11 | 08:55 10:34 | 34 ° 03.9205 ' 25 ° 01.0650 ' | 38300 | 99 / 1 | 100 | 342 | 24.6 |
| | 10/11 | 14:00 14:55 | 34 ° 09.3483 ' 24 ° 52.5356 ' | 46000 | 116 / 0 | 116 | 397 | 29.6 |
| | 11/11 | 06:50 07:49 | 34 ° 09.3491 ' 24 ° 52.5356 ' | 36000 | 101 / 0 | 101 | 356 | 28.6 |
| F/V SILVER LAGUNA | 06/11 | 20:10 21:10 | 33 ° 41.627 ' 26 ° 44.915 ' | 17400 | 43 / 26 | 69 | 239 | 26.1 |
| | 06/11 | 22:43 00:15 ⁽¹⁾ | 33 ° 36.021 ' 26 ° 57.743 ' | 13200 | 31 / 21 | 52 | 244 | 25.7 |
| | 07/11 | 12:35 19:00 | 33 ° 36.553 ' 26 ° 58.468 ' | 9500 | 20 / 20 | 40 | 238 | 25.6 |
| | 08/11 | 09:15 11:20 | 33 ° 37.661 ' 26 ° 56.070 ' | 8500 | 20 / 20 | 40 | 213 | 24.2 |
| | 09/11 | 01:40 05:35 | 33 ° 59.568 ' 25 ° 13.415 ' | 9000 | 20 / 20 | 40 | 225 | 23.4 |
| | 09/11 | 09:00 10:30 | 33 ° 59.853 ' 25 ° 14.537 ' | 5000 | 19 / 14 | 33 | 150 | 22.5 |
| | 09/11 | 11:55 18:15 | 34 ° 02.151 ' 25 ° 03.731 ' | 6500 | 20 / 0 | 20 | 325 | 29.9 |
| | 11/11 | 02:00 04:35 | 33 ° 59.864 ' 25 ° 14.534 ' | 9000 | 25 / 14 | 39 | 211 | 23.4 |
| | 11/11 | 06:49 10:05 | 34 ° 02.150 ' 25 ° 03.728 ' | 7500 | 24 / 0 | 24 | 313 | 29.4 |
| | 12/11 | 04:00 06:00 | 34 ° 09.339 ' 24 ° 25.917 ' | 7000 | 20 / 20 | 40 | 175 | 24.0 |
| | 12/11 | 17:15 20:30 | 34 ° 11.381 ' 24 ° 33.035 ' | 6000 | 25 / 20 | 45 | 130 | 23.5 |
| | 12/11 | 23:01 04:30 ⁽¹⁾ | 34 ° 09.318 ' 24 ° 52.744 ' | 2000 | 0 / 20 | 20 | 100 | 20.9 |
| | 13/11 | 17:00 21:00 | 33 ° 59.551 ' 25 ° 13.368 ' | 7000 | 23 / 20 | 43 | 160 | 23.7 |
| | 15/11 | 00:05 04:30 | 34 ° 09.347 ' 24 ° 52.537 ' | 7000 | 20 / 20 | 40 | 175 | 23.8 |
| Note (1) | | Session ended on following day. | | | | | | |

Appendix D

Extract from report on scientific research - 09 May to 20 May 2022 in First Closed Season

Hydro acoustic field work focusing on ground truthing and surveying of squid marks on the East Cape Coast inshore spawning grounds from R/V *Abyss*

1.0 Introduction

The primary objective of the closed season squid research exercise conducted on the eastern Cape Coast between 09 May and 20 May 2022 was to locate a sizeable squid aggregation at a depth between 20 and 40m in fairly close proximity to Cape St Francis which was to be used as a base for the scientific team participating in the exercise. Once such an aggregation had been located and confirmed (via jigging) a number of activities were to be carried out which included;

- Conduct small scale acoustic surveys to determine aggregation biomass.
- Perform target strength experiments where applicable.
- Investigate and attempt to sample marks occurring in the general vicinity of the aggregation to improve confidence in the allocation process applied when post processing survey data.
- Thoroughly document all sampling locations and the associated catch to enable correlation with the acoustic survey record.
- Calibrate the 38 kHz EK60 echo sounder on *R/V Abyss*.

The scientific team comprised Mr J. Rademan, Mr C. Erasmus and Ms D. Merkle from DFFE, and Mr. M Soule, an acoustics consultant from Fisheries Resource Surveys, Cape Town.

Acoustic targets were sampled using Yo-Zuri rods and various lures (Fig. 1)



Fig. 1: Selection of flies attached to the Yo-Zuri lines.

Figs. 2 and 5 to 8 from the Report show various acoustic targets which were sampled in this way, and Appendix D-1 the list and numbers of all species caught.

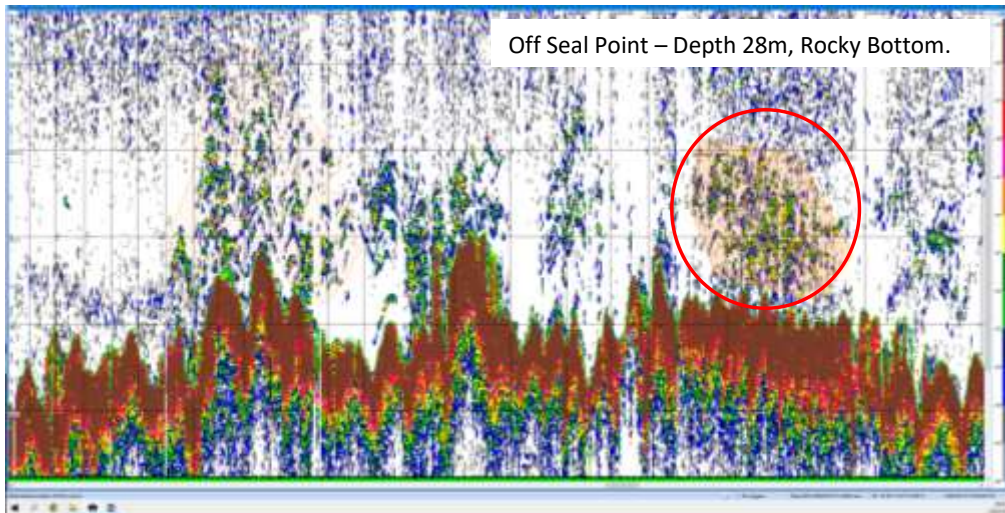


Fig. 2: Loosely aggregated targets above reef at a depth of 28m. The marked sector was targeted while drift sampling.

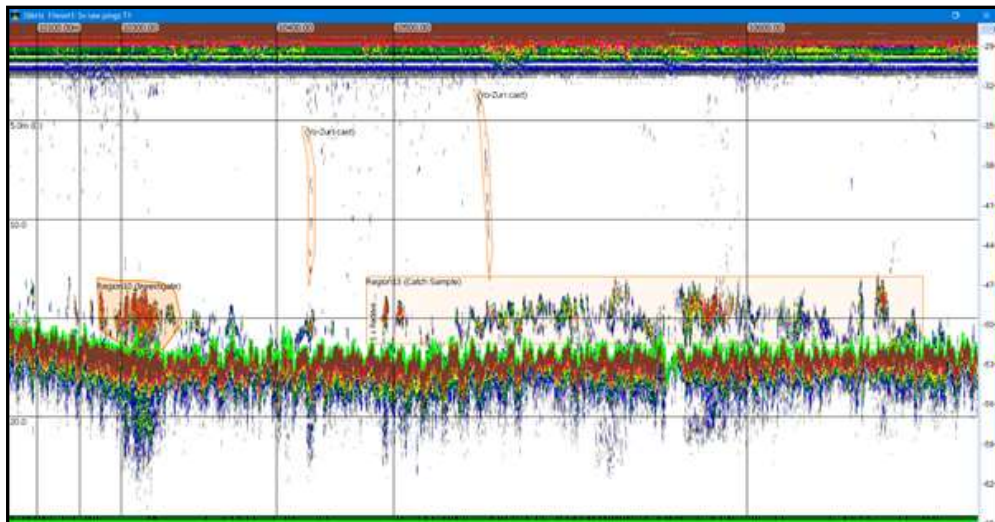


Fig. 5: (i) The dense bottom aggregation (LHS) is detected at survey speed (± 6 knots) on the echo sounder, (ii) *Abyss* breaks off from the survey, turns and drifts back over the mark (vertical lines are separated by 100m), (iii) Sampling of the layer on the bottom takes place using the Yo-Zuri rigs which can be seen on the echogram.

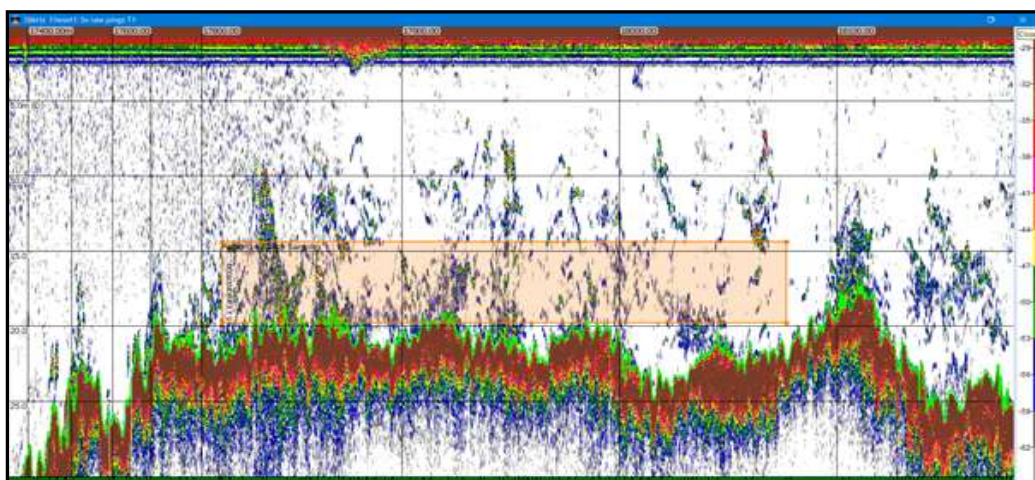


Fig. 6: (i) Loose shoaling fish on the bottom were detected at survey speed (± 6 knots) on the echo sounder(LHS), (ii) *Abyss* breaks off from the survey and drifts back over the mark (vertical lines are separated by 100m), (iii) Sampling of the layer on the bottom takes place using the Yo-Zuri rigs. Six Fransmadam's were caught while sampling along the bottom (depth ± 20 m) in the zone marked on the echogram.

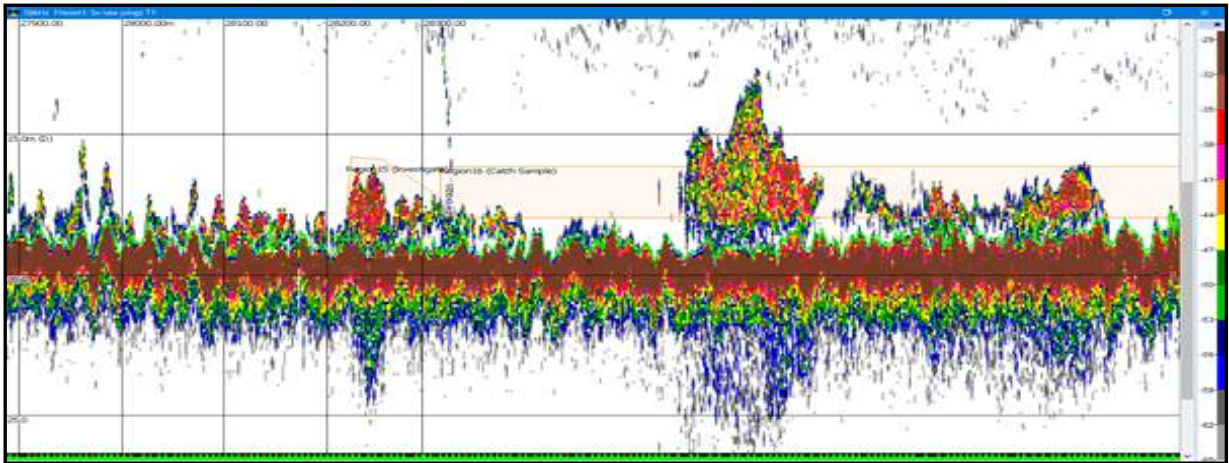


Fig. 7: Dense cloud like shoals on the bottom off the Kromme – possibly Piggys.

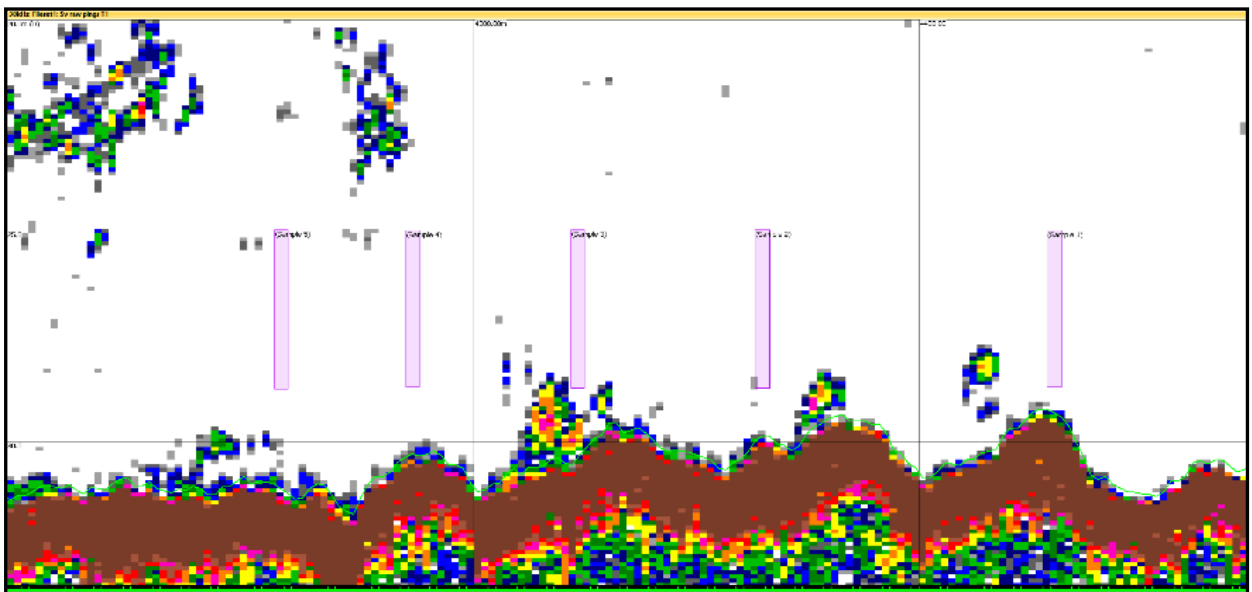


Fig. 8: Marks sampled during the final drift off the Kromme viewed at survey speed (± 6 knots) while following a reciprocal course about 15 mins. after the samples were initially taken. The catch consisted predominantly of 24-31 cm Carpenters (depth ± 30 m).

Summary

- No squid aggregations were detected during the surveys and drifts conducted between the 12th and 18th May, most probably because the squid are either not aggregated at this time of the year or are in deeper waters further offshore. This fact was reinforced by the data obtained from Balobi which showed that the squid landed in the last week of the open season (at the end of April) were caught under lights at night, most probably because the squid were dispersed during the daylight hours and not available to the catcher vessels.
- Drift experiments on marks of interest together with targeted sampling produced some interesting observations and will definitely contribute to a greater understanding of the diversity and behaviour of scatterers other than squid on the spawning grounds. Further experiments which improve on the methodology used here, for example conducting multiple drifts over specific marks of interest and increasing the size of the sample, should allow the species composition to be more accurately determined. Being able to confidently identify marks which at times may be confused with smaller less dense squid aggregations will hopefully improve the confidence in the allocation process applied during squid biomass surveys.
- *Abyss* was successfully calibrated on 17 May in excellent conditions off the Kromme. The results showed that the EK60 38 kHz split beam system has remained stable since it was last calibrated in April 2021 and continues to provide high quality data.

Appendix D-1
Numbers and size range of all species caught

| Species | Scientific name | N | Size range (cm) |
|---------------------------------|----------------------------------|----|-----------------|
| Fransmadam (Karel grootoog) | <i>Boopsoidea inornata</i> | 19 | 19.5 - 28.5 |
| Piggy | <i>Pomadasys olivaceum</i> | 16 | 11.0 - 21.0 |
| Carpenter | <i>Argyrozona argyrozona</i> | 15 | 20.0 - 31.0 |
| Red Tjor Tjor (Sand soldier) | <i>Pagellus natalensis</i> | 2 | 23.0 - 23.5 |
| Mackerel | <i>Scomber japonicus</i> | 2 | 42.0 , 43.0 |
| Cob | <i>Argyrosomus hololepidotus</i> | 1 | 30 |
| Elf | <i>Pomatomus saltator</i> | 1 | 12 |
| Barbel | <i>Arius feliceps</i> | 6 | 35.5 - 40 |
| Santer | <i>Cheimerius nufar</i> | 5 | 15.0 - 23.0 |
| Strepie (Karanteen) | <i>Sarpa Salpa</i> | 3 | 21.0 - 25.5 |
| Maasbanker (Horse Mackerel) | <i>Trachurus capensis</i> | 2 | 22.5 |
| Red Roman | <i>Chrysoblephus laticeps</i> | 2 | 27.0 |
| Redeye | <i>Etrumeus whiteheadi</i> | 2 | 22.5 - 25.0 |
| Blue Hottentot | <i>Pachymetopon aeneum</i> | 1 | 29.5 |
| TOTAL | | 77 | |