



Improving Knowledge of Dugong Life History Using Surrogate Data

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**Marine and Tropical Science Research Facility
Projects 2007-08**

FINAL REPORT

Project 1.4.1 Objective A:

Condition, status and trends and projected futures of the dugong in the Northern Great Barrier Reef and Torres Strait; including identification and evaluation of the key threats and evaluation of available management options to improve its status.

Complementary Objective C.

Improving knowledge of dugong life history.

Investigators:

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Work Undertaken to Date Enabled by Supplementary Funding from GBRMPA

- (1) The Growth Layer Groups of one tusk from each of 20 dugongs from Torres Strait were examined using a range of techniques to investigate whether they provide an unambiguous record of: a) early growth; b) the age of sexual maturity in males and females, and c) calving events. The life history of each of the dugongs was inferred from carcass analysis.
- (2) The aerial survey records collected in Australia since the mid 1970s were used to quantify spatial and temporal patterns of fecundity using percentage calf counts as a surrogate measure of dugong fecundity. These data were then modelled to determine if changes in dugong fecundity are associated with weather indices such as the SOI, NINO 3.4 or records of rainfall.

Results

- (1) The spacing in the GLGs of the tusks of both female and male dugongs from Torres Strait does not show a distinct pattern that can unequivocally be attributed to life history events such as the onset of sexual maturity or calving. Plausible scenarios can be developed to explain the pattern of GLG deposition in individual dugongs for which life history information is available from carcasses. However, these patterns are too uncertain for the pattern of GLG deposition in the tusk of a dugong from a tropical region such as Torres Strait to be used as an unambiguous record of its life-history.
- (2) The temporal and spatial differences in dugong fecundity measured by the proportion of calves seen during aerial surveys are large. In regions that were comprehensively surveyed, the proportion of calves ranged from 0.002 in the Northern Great Barrier Reef in 1978 to 0.22 in Hervey Bay in 1988 and 1992.

There was a significant negative relationship between the proportion of calves and the wet season rainfall, the wet season rainfall anomaly and the SOI and a positive relationship between the proportion of calves and the wet season NINO3.4, each lagged by two years in the Northern Great Barrier Reef region.

In the Western Australian survey region there was a significant negative relationship between the proportion of calves and the SOI and a significant positive relationship between the proportion of calves and NINO3.4, each lagged by two years; but the relationship with rainfall or rainfall anomalies could not be tested because of the low variation in rainfall. In the Moreton Bay region, the proportion of calves positively covaried with rainfall lagged by two years and negatively covaried with rainfall and rainfall anomalies each lagged by three years (which may have been a consequence of the two-year positive relationship), but there was no relationship with SOI or NINO 3.4. The results for Western Australia and Moreton Bay are tentative as they were not robust over all covariates. It is possible that similar relationships exist for other regions and that the data are not yet adequate to confirm them statistically (Type 2 error).

- (3) The negative impact on dugong fecundity of the loss of coastal seagrass associated with exceptionally high rainfall and other extreme weather events is of major concern when considering the impact of climate change on dugongs.

Recommendations

- (1) That the use of dugong tusks as a record of life history events be restricted to age determination.
- (2) That the proposed Masters project to further examine dugong tusks as a record of life history events not be carried out because this project demonstrates that dugong tusks from Torres Strait are an ambiguous record of such events.
- (3) That the data on the proportions of calves seen on dugong surveys be further analysed in collaboration with meteorologists in order to better capture any impact of pulsed turbidity events.
- (4) That future assessments of the impact of pulsed turbidity events associated with Climate Change for the GBRWHA include the negative impact of such events on dugong fecundity.
- (5) That the dugong stranding database be examined to determine whether the number of deaths attributed to natural causes is correlated with broad-scale sources of inter-annual variability in weather and climate such as changes in rainfall, SOI and NINO 3.4.

Outcomes/Objectives

Objective 1:

Provide a final report on the pilot study conducted to examine if the spacing and elemental context of the growth layer groups in the tusks and ear bones of dugongs

collected from the Torres Strait reflect the life history events in animals known reproductive histories.

This objective has been achieved for tusks. It was not possible to undertake specimen preparation for the ear bones within the time frame of the project because the time required to decalcify dugong ear bones is many months. However, we regard it as extremely unlikely that the ear bones would have yielded more information than the tusks.

Objective 2:

Include in the final report the spatial and temporal patterns of dugong fecundity using percentage counts as a surrogate measure of dugong fecundity from the aerial survey records of dugong distribution and abundance collected by James Cook University for the east coast of Queensland and Torres Strait since the 1980s.

This objective has been achieved.

Objective 3:

If possible, include in the final report additional data on: dugong life history parameters such as age at sexual maturity, calving and duration of lactation and the way they change through time at a population level; and periods of nutritional stress during low food abundance.

This objective has been achieved for the relationship between fecundity and weather only. It was not possible to undertake the remainder of this part of the project because the tusks were not sufficiently informative.

Appropriateness of the Approaches used in the Development and Implementation of the Project

Appropriate

Communicating the Results to Policy Makers and Stakeholders

Dr Kirsten Dobbs and Ms Stephanie Lemm GBRMPA have been briefed verbally by Professor Helene Marsh.

Transferability of the Research

The relationship between rainfall and dugong fecundity should be explored for other areas in the dugong's range and used in the development of models to inform policy about the levels of human-induced mortality that are likely to be sustainable.

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Executive Summary

- (1) The Growth Layer Groups of one tusk from each of 20 dugongs from Torres Strait were examined using a range of techniques to investigate whether they provide an unambiguous record of: a) early growth; b) the age of sexual maturity in males and females; and c) calving events. The life history of each of the dugongs was inferred from carcass analysis.
- (2) The spacing in the GLGs of the tusks of both female and male dugongs from Torres Strait does not show a distinct pattern that can unequivocally be attributed to life history events such as the onset of sexual maturity or calving. Plausible scenarios can be developed to explain the pattern of GLG deposition in individual dugongs for which life history information is available from carcasses. However, these patterns are too uncertain for the pattern of GLG deposition in the tusk of a dugong from a tropical region such as Torres Strait to be used as an unambiguous record of its life-history.
- (3) The aerial survey records collected in Australia since the mid 1970s were used to quantify spatial and temporal patterns of fecundity using percentage calf counts as a surrogate measure of dugong fecundity. These data were then modelled to determine if changes in dugong fecundity are associated with weather indices such as the SOI, NINO 3.4 or records of rainfall.
- (4) The temporal and spatial differences in dugong fecundity measured by the proportion of calves seen during aerial surveys are large. In regions that were comprehensively surveyed, the proportion of calves ranged from 0.002 in Northern Great Barrier Reef in 1978 to 0.22 in Hervey Bay in 1988 and 1992. There was a significant negative relationship between the proportion of calves and the wet season rainfall, the wet season rainfall anomaly and the SOI and a positive relationship between the proportion of calves and the wet season NINO3.4, each lagged by two years in the Northern Great Barrier Reef region. In the Western Australian survey region there was a significant negative relationship between the proportion of calves and the SOI and a significant positive relationship between the proportion of calves and NINO3.4, each lagged by two years; but the relationship with rainfall or rainfall anomalies could not be tested because of the low variation in rainfall. In the Moreton Bay region, the proportion of calves positively covaried with rainfall lagged by two years and negatively covaried with rainfall and rainfall anomalies each lagged by three years (which may have been a consequence of the two-year positive relationship), but there was no relationship with SOI or NINO 3.4. The results for Western Australia and Moreton Bay are tentative as they were not robust over all covariates. It is possible that similar relationships exist for other regions and that the data are not yet adequate to confirm them statistically (Type 2 error).

- (5) The negative impact on dugong fecundity of the loss of coastal seagrass associated with exceptionally high rainfall and other extreme weather events is of major concern when considering the impact of climate change on dugongs.

Recommendations

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1. Introduction

The dugong, the only extant member of the family Dugongidae, is listed as vulnerable at a global scale by the International Union for Conservation of Nature (IUCN 2007). In Australia and Queensland, the dugong is protected by the *Environment Protection and Biodiversity Conservation Act 1999* (Cth) and the *Nature Conservation Act 1994* (Qld), respectively. Dugongs have a wide global range which spans at least 48 countries (Marsh et al. 2003 and unpublished). Australian waters support more dugongs than those of any other country. As the only developed country with a significant dugong population, Australia is internationally expected to play a pivotal role in dugong conservation. The significance of the Great Barrier Reef World Heritage Area (GBR WHA) as a feeding ground for dugongs is an explicit reason for that Area's World Heritage Listing (GBRMPA 1981). In Torres Strait, the customary dugong fishery is recognised by the international treaty between Australia and Papua New Guinea.

Dugongs are long-lived animals with a slow reproductive rate, long generation time and high investment in each offspring (Marsh et al. 1984 a, Boyd et al. 1999). These factors make them susceptible to anthropogenic threats, particularly anthropogenic mortality, as the dugong's life history makes the species particularly sensitive to changes in adult survivorship. Population models also demonstrate that the rate of increase of a dugong population is sensitive to changes in fecundity (Heinsohn et al. 2004).

Cryptic marine mammals such as dugongs are notoriously difficult to study. These difficulties are exacerbated when such animals usually occur in: (1) relatively large numbers (thousands), (2) turbid water, (3) relatively remote areas (Marsh et al. 2003), and (4) are highly mobile (Sheppard et al. 2006). These features make mark-recapture studies logistically impossible throughout much of the dugong's range in Australia, even though they have potential in Moreton Bay, the only urban area which supports dugongs in a clear water area that is relatively isolated from the remainder of the range (Lanyon et al. 2002). As a result of these logistical challenges, almost all information on dugong life history originates from carcass analysis (Marsh 1980, Marsh et al. 1984a, Boyd et al. 1999, Kwan 2002, Marsh and Kwan in press).

Growth Layer Groups (GLGs) in dugong tusks have been used to estimate age (Marsh 1980). In conjunction with the examination of reproductive specimens, these estimates of age have been used to estimate life history parameters such as the pre-reproductive period and the calving interval, both of which show considerable variation in both space and time (Boyd et al. 1999, Kwan 2002, Marsh and Kwan in press). Marsh and Kwan (in press) demonstrated that the life history and reproductive rate of female dugongs are adversely affected by seagrass loss, which in turn, can result from natural environmental factors such as light deprivation resulting from increases in turbidity resulting from prolonged flooding (Preen and Marsh 1995, Preen et al. 1995) or sediment resuspension during prolonged periods of high winds (Saint-Cast in press). Green turtle life history traits such as growth and breeding rates also vary with environmental factors (Limpus and Nichols 1988; Chaloupka et al. 2004). For example, the number of green turtles that breed each year increases two years following an El Niño episode (Limpus and Nichols 1988). El Niño episodes, which are associated with negative values of the Southern Oscillation Index (SOI) and

positive values of the NINO 3.4 sea surface temperature index, are usually accompanied by sustained warming of the central and eastern tropical Pacific Ocean, a decrease in the strength of the Pacific Trade Winds, and a reduction in rainfall over eastern and northern Australia. NINO 3.4 is one of the regions in the equatorial Pacific important for monitoring and identifying El Nino and La Nina. The sea surface temperature variability in this region has a strong effect on rainfall variation in the western Pacific. Calves are recorded separately from adults during dugong aerial surveys. The time series of dugong aerial surveys conducted in northern Australia since the 1970s (Table 1) provide an extensive record of variation in dugong fecundity over huge temporal and spatial scales. The potential of the data from these surveys to reveal whether dugong fecundity also varies with broad-scale sources of inter-annual variability in weather and climate such as changes in the Southern Oscillation Index and the NINO 3.4 sea surface temperature index has not been investigated.

The Potential Biological Removal method (Wade 1998) has been used to estimate sustainable anthropogenic mortality from all causes for dugongs in various regions including the GBRWHA and Torres Strait (Marsh et al. 2004a, 2006 and 2007). This technique requires an accurate and precise knowledge of dugong life history parameters based on empirical data. At present the information on age parameters such as age of first reproduction and calving rate (both of which vary in space and time as discussed above) are based on relatively small samples because of the need to collect reproductive specimens as well as tusks for age determination to measure these parameters.

Unlike dugongs, Florida manatees do not have tusks. Marmontel (1995) and Marmontel et al. (1996) used the GLGs in manatee ear bones for age determination in a technique analogous to that used by Marsh (1980) for dugong tusks. The patterns of inter-annual variation in the width of the GLGs in their ear bones have subsequently been verified as proxies of life history information in individual Florida manatees with well-documented life histories. Pitchford and Rommel (2002) compared the timing of events from an individual manatee's life history with the timing of inter-annual variations in growth observed in the measurements of GLGs. Their results indicated that: (1) a consistent and relatively large amount of growth was observed during the first three years for all individuals examined, (2) all individuals had a distinct change in growth rate between ages three and five, consistent with the documented timing of sexual maturity and field observations of the manatees examined for this study, and (3) after age five, each manatee had intervals of inconsistent growth which may reflect individual life history events. They considered that GLG spacing could be a useful method for obtaining life history information of manatees and that growth-layer intervals could be used for both population level assessments by examining long-term trends in age of sexual maturity, and for individual assessments by identifying short-term biological and environmental events.

The potential to use the GLGs in dugong tusks in this manner to increase the sample size on which life history parameters are based has considerable advantages, particularly because of the inherent difficulties in obtaining such information from either: (1) carcasses that are to be used for meat by Traditional Owners when researchers are not present at the butchering; or (2) decomposed carcasses. If dugong tusks could be used as a record of major life history events, the information could be used both in population models and in public education to provide information to

individual hunters on their catch. Similarly, calf counts obtained during aerial surveys for dugongs are potentially useful proxies of dugong fecundity. If they vary in association with extreme weather events, large scale changes in fecundity, which also affect the rate, could be predicted on the basis of weather records in the same manner as temporal variation in green turtle nesting is predicted (Limpus and Nichols, 1988, 2000).

This project thus had two aims:

- (1) to determine whether dugong tusks provide an unambiguous record of: a) early growth; b) the age of sexual maturity in males and females; and c) calving events by examining the tusks of dugongs for which life history information could be inferred from carcass examination.
- (2) to examine the aerial survey records collected in Australia since the late 1970s to quantify spatial and temporal patterns of fecundity using percentage calf counts as a surrogate measure of dugong fecundity to determine if changes in dugong fecundity are associated with weather indices such as the SOI or NINO 3.4 or rainfall records.

2. Materials and Methods

2.1 Dugong Tusks as a Record of Life History Events

2.1.1 Source of Material

The tusks (second incisors) of 92 female and 51 male dugongs of a range of ages and reproductive states collected by Kwan (2002) from the dugong fishery conducted at Mabuiag Island in Torres Strait were available for analysis. The date of death of each animal was known. Life history information was available for most of the animals from gross and histological examination of their gonads and the mammary glands of female dugongs.

The tusks were strategically sub-sampled to enable tusks to be selected from animals with a comprehensive and unambiguous record of life history events. Twenty dugong tusks were examined, 16 from female dugongs and four from male dugongs (Table 2).

2.1.2 Growth Layer Groups (GLGs) in Tusks

The postnatal dentine is deposited in a dugong tusk in a prolonged series of coaxial cone-shaped increments. Each GLG consists of two zones (A and B in Marsh 1980). Zone A forms the ridge in acid-etched tusks; Zone B the groove. Using the proportional increment method, Marsh (1980) examined the thickness of the Zone being laid down at the time of death and found that: (1) Zone B of the GLG was almost always formed between July and October, and (2) the thickness of Zone A increased from October to September. Both these results suggest that one GLG is deposited per year. The width of the GLGs decreases in older animals. The inclination of the GLGs from the long axis of the tooth also increases with time, resulting in the later layers in very old teeth being almost perpendicular to the long axis of the tooth.

2.1.3 Methodologies trialled to study the Growth Layer Groups in Dugong Tusks

Marsh (1980) trialled several methods of preparing dugong tusks for age determination including etched half-teeth, stained transverse thick (500µm) and thin (30-80µm) sections and microradiographs of transverse thin sections (280-600µm). She found that the numbers of GLGs revealed by each method were similar and suggested that acid-etched half teeth were most appropriate for counting GLGs for age determination. Marsh (1980) reported that the GLGs in the tusks of dugongs from the areas in her study (mainly Townsville and Mornington Island) were extremely clear and usually easy to count. She noted that Mitchell (1978) observed that tusks from Torres Strait were difficult to read because they had more marked accessory layering than teeth from Townsville. The more marked accessory layering in dugong tusks from Torres Strait (and Banka Island in Indonesia) was confirmed by participants at an Age Determination Workshop in 1979 (Marsh and Kasuya 1981). Workshop participants concluded that the degree of accessory layering is inversely correlated with latitude, perhaps as a result of tropical environments being less seasonal than temperate ones.

We needed to develop methods to enable us to measure the width of tusk GLGs, rather than simply count them as Marsh (1980) had done. We spent considerable effort investigating a range of methods, most of which proved unsuitable (Table 3). Previous studies have used transverse thin sections to show up GLGs in the teeth of marine mammals but dugong tusks exhibit dramatic axial growth in both sexes, making transverse sections useless for age determination based on dentinal layering. For our purposes, the most suitable method was to measure the GLGs on high resolution digital photographs of acid-etched half-tusks using image processing techniques as outlined below.

2.1.4 Sample Preparation

Acid- etching to Highlight Growth Layer Groups

Kwan (2002) cut the tusks in half longitudinally in the mesiodistal plane with a 20cm carbon steel blade. We used a half tusk from each individual that had not been previously acid-etched. The cut surface was polished using progressively finer grades (180, 240, 800 and 1200 grit) of wet and dry sandpaper. Each tusk was then etched in 5% formic acid (*sensu* Pierce and Kajimura 1980) for 6-10 hours. The changes in each tusk was individually monitored during the etching process by removing it from the acid bath every two to three hours, briefly rinsing it in running water, placing it in acetone for several minutes and then allowing it to air dry for at least ten minutes. This procedure permitted accurate determination of the degree of etching achieved and prevented over-etching. After the level of etching was considered sufficient, each tusk was rinsed thoroughly in water for at least five minutes before being left to soak in a tray under gently running water for a further 12 hours. The tusk was then air dried for one to two days. The tusks must be rinsed thoroughly before final drying to prevent gradual deterioration of the etched surface after it is dried from the residual acid (Pierce and Kajimura 1980; Kwan 2002).

High Resolution Digital Photography

Digital photographs were taken of each acid-etched half tusk using a Fuji FinePix S2 Pro camera, ISO 100, focal length 70mm, exposure 1/25s @ f/6.7 with a flash (Single Bowens studio flash unit). The contrast and brightness of the images were transformed in Photoshop to enhance the layering. Each photograph included a 10mm scale bar to enable measurements in the image analysis software to be calibrated as outlined below.

2.1.5 Measuring GLGs

The digital image of each tusk was viewed in ArcGIS9. Lines were drawn on the image to delineate each ridge and groove within each GLG. The widths of the GLG ridge and groove changes from the centre line to the edge of the tusk and therefore the average of several measurements was used to determine the width of each ridge or groove. Measurements were taken along the edge of the ridge or groove at 5mm increments from the edge of the tusk. A perpendicular line was drawn between the lines delineating each ridge and groove using the sketch tool in ArcGIS9 (Figure 1). The lengths of these perpendicular lines, calculated by ArcGIS9, were exported into Excel for graphing to compile a time series of widths of GLGs (Appendix 1). The first three or four dentinal layers were much more difficult to measure than the layers formed subsequently. Therefore each time series was started from the third GLG.

2.1.6 Assessing the Repeatability and Reliability of Measurements

The measurement of a structure may vary depending on the equipment/software used to make the measurements, the person taking the measurements or within person variation across time. Two observers (JG and RH) measured the widths of the GLGs independently after examining several tusks together to decide on an agreed protocol.

The repeatability and reliability of the measurements of the widths of GLGs in tusks were calculated to assess the differences in measurements between the two persons taking the measurements (between observer differences) and the differences in measurements made by the same observer through time (within observer differences).

For each of four tusks (MD56, MD 21, MD41 and MD161), the two observers each independently measured the widths of the same five GLGs in each tusk on four occasions to assess measurement repeatability both between and within observers. The time between measurements was long enough for successive observations by the same observer to be regarded as independent within observer measurements.

We used the method of Lessells and Boag (1987) to estimate repeatability as the intra-class correlation coefficient (Sokal and Rohlf 1995) based on variance components derived from a one-way analysis of variance. Repeatability, the proportion of variation in the sample among observers varies from 0 to 1. A high value of this coefficient means that most of the variation in the sample is among observers. A value of unity would indicate that all of the variance is among observers (i.e. there is no variance within observers). Reliability was calculated in the same way. The intra-class correlation coefficient, r is given by:

$$r = S^2_A / (S^2 + S^2_A)$$

where, S^2_A is the among-observer variance component and S^2 is the within-observer variance component. The variance components are calculated from the mean squares in the analysis of variance as:

$$S^2 = MS_w$$

and

$$S^2_A = (MS_A - MS_w)/n$$

where n is the sample size per observer. A worked example of the calculation is provided in Appendix 2.

The repeatability and reliability of the GLGs counts were also evaluated for each of the four tusks because the accessory layering made it very difficult to ensure consistency in interpreting some structures. The proportion of times an observer measured a different structure was calculated from the five GLGs measured in each of the four tusks for Observer 2 and in each of two tusks for Observer 1.

2.1 7 Aligning Tusk Measurements and Life History Information

As detailed in Appendix 1, the pattern of the spacing of GLGs in each dugong tusk was presented graphically and considered in the context of the available information on that animal's life history. The analysis makes the following assumptions:

- (1) that the layering in dugong tusks is a reliable record of growth rhythms (Kelvezal 1980);
- (2) that one GLG is laid down per year, with Zone A in the Summer and Zone B in the winter (Marsh 1980);
- (3) that Zone A is a record of growth and fat deposition;
- (4) that Zone B represents a period of growth retardation, or loss in body condition;
- (5) that dugongs undergo a period of fat deposition before conception and during pregnancy (Kwan 2002; Marsh and Kwan in press);
- (6) that placental scars are a reliable long-term index of birth events (Marsh et al. 1984 b).

2.2 Temporal Variation in Dugong Fecundity as revealed by Calf Counts during Aerial Surveys throughout Northern Australia between 1974 and 2006.

Figure 2 and Table 1 list the dates and locations for the aerial surveys from which the data on dugong calf counts were extracted. Figure 2 shows the range of the dugong in Australia, illustrating place names mentioned in Table 1. The number of dugong calves and the total count of dugongs in each survey year were summed over regions within each of seven locations: (1) Western Australia (Shark Bay, Ningaloo Reef and Exmouth Gulf), (2) Gulf of Carpentaria (various sections including the entire gulf, the Queensland coast, the Northern Territory coast, Cape York to Weipa, and Karumba to Bayley Point), (3) Torres Strait, (4) Northern Great Barrier Reef (north of Cooktown to Hunter Point), (5) Southern Great Barrier Reef (Cooktown to the southern border of the GBR Marine Park), (6) Hervey Bay, and (7) Moreton Bay. Only one survey of the northern coast of the Northern Territory was available so it was not included in the analyses.

Generalised linear models (GLM) (Wood 2006) were used to analyse the relationship between the proportion of dugong calves and four environmental covariates: (1) total wet season (October – March, *sensu* Lough 1997) rainfall averaged over the coastal weather stations in the survey region or subregion (Table 4), (2) wet season rainfall anomaly (difference between wet season rainfall for a survey region (subregion) and the 30 year average for that region (subregion)), (3) the November to February Southern Oscillation Index (SOI) and (4) the November to February NINO 3.4 sea surface temperature index (*sensu* Cheal et al 2000). Lagged relationships with each covariate were assessed for each of one to four years prior to the survey year. **Lagged relationships were expected because of the long reproductive cycle of the dugong (Boyd et al. 1999).** The models estimated the proportion of calves in each location and the linear relationship between the proportion of calves and each external covariate (at each lag). Exploratory analyses did not find evidence for nonlinear relationships. GLMs were fitted with a logit link function and binomial variance, and the proportion of calves was weighted by the total number of animals. All analyses were done using the R statistical software system (R Development Core Team 2007).

Analyses were also performed on the east coast only data from the three locations with multiple regions within each location. Generalised linear mixed models (Breslow and Clayton 1993, Bates and Saikat 2004) were used to account for additional random variation among blocks within regions. Otherwise, model specification was the same.

3. Results

3.1 Growth Layer Groups in Tusks

3.1.1 Description of the Changes in the General Patterns in the Spacing of Growth Layer Groups with Increasing Age

The first three growth layers were generally very difficult to measure in the tusks of both sexes. The boundaries of the growth layers could not be identified with certainty because of the pronounced accessory layering. As expected, the layering indicated that growth was rapid to about age four in most individuals but the patterns in the spacing of GLGs in tusks did not give a clear indication of when early growth stops, particularly in males. (Note: This result is not always depicted in the graphs in Appendix 1 because we generally could not measure the early GLGs because of the accessory layering).

3.1.2 An Assessment of the Potential for Patterns in Spacing of Growth Layer Groups to mark Life History Events or Environmental Factors.

With knowledge of the life history obtained from carcass analysis, plausible scenarios were developed to explain the patterns in spacing of growth layer groups in the 20 dugongs studied in detail (see Appendix 1). However, these patterns were open to alternative interpretations and for most females the tusks suggested multiple life history scenarios of uncertain validity. If males experience a pre-pubertal growth spurt, the tusks may provide a record of sexual maturity in males but this finding is of doubtful validity without additional data. We conclude that the patterns of layering in

the tusks of dugongs from Torres Strait are not clear enough to be used as a reliable record of life history events in animals of unknown life history.

3.1.3 Assessing the Repeatability and Reliability of Measurements

The measurements made by each of the two observers were reliable and repeatable because values of r were relatively small indicating that for each observer there was little error in their repeated measurements of GLGs in the same tusk. Error rates ranged from 3.4% to 24.5% (Table 5) which is excellent for macroscopic measurement of structures <1mm. Similarly, for Time 1 there was little error in the measurements of GLGs in the same tusk between the two observers. Error rates ranged from 9.26% to 24.8% for these tests (Table 6).

The counts of GLGs were also reliable and repeatable. The error rates in counting GLGs were 22.4% ($F = 0.08$) for Observer 1 and 22.5% ($F = 0.15$) for Observer 2. The error rate between observers for Time one was 23.5% ($F = 0.05$).

A different structure was measured by Observer 1 in 10% of cases (four times out of a possible 40 times – 5 GLGs x 2 tusks x 4 times) and by Observer 2 in 13.75% of cases (11 times out of a possible 80 times – 5 GLGs x 4 tusks x 4 times).

The rate of error in the measurements of GLGs within and between observers was relatively small (<25%) and unlikely to have greatly affected our assessment of the patterns of spacing of GLGs.

3.2 Dugong Fecundity

In regions that were comprehensively surveyed, the proportion of calves ranged from 0.002 in Northern Great Barrier Reef in 1978 to 0.22 in Hervey Bay in 1988 and 1992. Hervey Bay had the largest range of values at a single location because the proportion of calves in 1994 was only 0.015 (Figure 3). The average proportion of calves over all years in each location ranged from 0.050 in Moreton Bay to 0.129 in Torres Strait (Table 7). The relationship between the rainfall lagged by zero to three years inclusive and the proportion of calves is presented in Figure 4 for the whole data set; the corresponding relationship for rainfall anomalies in Figure 5, SOI in Figure 6 and NINO 3.4 in Figure 7.

3.2.1 Analyses Based on All Locations

The calf count data were collected in different years within each location (Table 1), and over time the blocks sampled within each location also changed. These limitations excluded the comparison of calf proportions between locations over time. The different sampling years also resulted in different distributions of values of the external covariates (total wet season rainfall, wet season anomaly, SOI and NINO 3.4). For example, in some locations the range of wet season rainfall values observed over survey years (or some lag thereof) were large, whereas in other locations only small variations in rainfall were observed among survey years (or some lag thereof). Consequently, overall estimates of the relationships between the covariates and calf proportions were not possible, because apparent relationships may be dominated by

among location differences in the covariate. Figure 4a highlights this situation: the black points (Western Australia) only occur at relatively low rainfall values, as do the yellow (Moreton Bay) and pink (Hervey Bay) points. Therefore, models were fitted with varying location differences and varying slopes of the calf proportion-environment relationship between locations.

These location differences in the variation in summer rainfall values in the survey years (or lags thereof) limited the power of the analysis to detect any relationship between wet season rainfall and rainfall anomalies (or lags thereof) and the proportion of calves. There was no variation in rainfall among years in the Western Australia location and therefore this region was excluded from that analysis. **There was a significant relationship for the Northern Great Barrier Reef, where the proportion of calves negatively covaried with rainfall and rainfall anomaly lagged by two years (e.g. Wald = 2.04, $P = 0.047$ for rainfall; Figure 4c).** Increases in rainfall of 0.1m in the wet season two years before surveys decreased the odds of dugong calves by 7% (95% CI = 0.3%, 14.3%; Figure 8). In contrast, the proportion of calves positively covaried with rainfall lagged by two years in Moreton Bay (Wald = 2.9, $P = 0.006$; Figure 4c). Increases in rainfall of 0.1m in the wet season two years before surveys increased the odds of dugong calves by 22.2% (95% CI = 7.2%, 39.3%; Figure 8). The counterintuitive result for Moreton Bay may be associated with the negative relationship between rainfall and calf proportions was observed at a 3-year lag for this location (Wald = 3.02, $P = 0.004$; Figure 4d). Increases in rainfall of 0.1m in the wet season three years before surveys decreased the odds of calves by 25.1% (95% CI = 8.2%, 44.7%; Figure 8). No other relationships were observed with rainfall at other lags. The results were equivalent for the rainfall anomalies (Figure 5 and Figure 9).

Consistent with the results for rainfall, the proportion of calves negatively covaried with SOI lagged by 2 years in the Northern Great Barrier Reef (Wald = 3.1, $P = 0.003$; Figure 6c). One unit increases in the mean wet season SOI two years before surveys decreased the odds of dugong calves by 3.8% (95% CI = 1.4%, 6.4%; Figure 10). The proportion of calves also negatively covaried with SOI lagged by two years in Western Australia (Wald = 3.51, $P = 0.001$; Figure 6c). One unit increases in the mean wet season SOI two years before surveys decreased the odds of dugong calves by 6.9% (95% CI = 3.0%, 10.9%; Figure 10).

Additionally, the proportion of calves positively covaried with NINO3.4 lagged by two years in the Northern GBR (Wald = 3.2, $P = 0.002$; Figure 7c). Increases in the mean wet season NINO3.4 by 10% two years before a survey increased the odds of dugong calves by 2.6% (95% CI = 0.3%, 23.5%; Figure 11). The proportion of calves also positively covaried with NINO3.4 lagged by two years in Western Australia (Wald = 3.3, $P = 0.002$; Figure 7c). Ten percent increases in the mean wet season NINO3.4 two years before surveys increased the odds of dugong calves by 0.3% (95% CI = 0.01%, 10.1%; Figure 11).

3.2.2 Analyses based on East Coast Locations only: (1) Northern Great Barrier Reef, (2) Southern Great Barrier Reef, and (3) Torres Strait.

The annual mean proportion of calves and 95% confidence intervals for the east coast survey regions which were sufficiently large as to be divided into subregions are presented in Table 8 and Figure 12. Results of the GLM analysis were consistent with

those from the aggregated data from these locations. The proportion of calves negatively covaried with rainfall and rainfall anomaly lagged by two years only in the Northern Great Barrier Reef (e.g. Wald = 2.6, $P = 0.014$ for rainfall). Increases in rainfall of 0.1m in the wet season two years before surveys decreased the odds of dugong calves by 8.6% (95% CI = 2.0%, 15.6%; Figure 13). The results were equivalent for wet season anomalies (Figure 14). The proportion of calves negatively covaried with SOI lagged by two years in the Northern Great Barrier Reef (Wald = 4.1, $P = 0.0001$). One unit increases in the mean wet season SOI two years before surveys decreased the odds of dugong calves by 1.0% (95% CI = 0.1%, 9.0%; Figure 15). The proportion of calves positively covaried with NINO 3.4 lagged by two years in the Northern GBR (Wald = 4.1, $P = 0.0002$). One unit increases in the mean wet season NINO3.4 two years before surveys increased the odds of dugong calves by 54.3% (95% CI = 25.1%, 90.2%; Figure 16)

4. Discussion

4.1 Growth Layer Groups in Tusks

Klevezal (1980) has shown experimentally that layers in the hard tissues of mammals are a record of the growth rhythms of individuals. However, this rhythm seems to be less distinct in dugong tusks from Torres Strait than in the ear bones of Florida manatee which live in a much more seasonal environment (Pitchford and Rommel 2002). The layering in dugong ear bones is less clear than that of tusks (Marsh unpublished information) and the time frame of this study did not allow us to investigate them further because months of decalcification required before histological sections can be prepared from ear bones.

The spacing in the GLGs in the tusks of dugongs suggested relatively rapid growth up to about age four years but large individual variation in the pattern and timing of such growth. This result is consistent with the dugong growth curve generated by Marsh (1980) who plotted the relationship between body length at death and the number of growth layers in the tusk. However, unlike Florida manatees, which exhibited a consistent and relatively large amount of growth observed during the first three years for all individuals (Pitchford and Rommel 2002), patterns in the spacing of GLGs in dugong tusks did not give a clear indication of when early growth stops, particularly in males. Klevezal (1980) predicted that in long-lived animals such as marine mammals, growth in the first few years of life will persist for as long as is permitted by external factors. It is not surprising that the rhythm of growth is less predictable in the tropical waters of Torres Strait than in the more temperate environment of Florida.

Florida manatees exhibited a distinct change in growth rate between three and five years, consistent with the documented timing of sexual maturity in manatees (Boyd et al. 1999). In contrast, dugongs did not show a consistent distinct change in growth rate at a particular age. This result is likely to result from the fact that female dugongs exhibit considerable variability in age at sexual maturity among individuals, within populations at different times and possibly among populations (Marsh 1995; Boyd et al. 1999; Kwan 2002). Female dugongs become sexually mature between six and 18 years (Marsh 1995, Boyd et al. 1999, Kwan 2002, Marsh and Kwan in press).

On the basis of carcass examination, Kwan (2002) concluded that the female dugongs that formed the basis of our sample typically conceived soon after they became sexually mature. If a distinct change in the pattern of spacing in GLGs in the tusks indicated the onset of sexual maturity in female dugongs, then the tusks of those dugongs in our sample that were pregnant with or suckling their first calf when they died should show an increase in the width of Zone A (the ridge) in a GLG in the year they died or the preceding two years. Although the tusks of all four dugongs in these categories (MD19, MD58, MD 101 and MD122) suggested superior growth/fat deposition coincident with presumed pregnancy, these patterns were not distinct enough to be interpreted as a certain record of the onset of sexual maturity.

After age five, manatees showed intervals of inconsistent growth which Pitchford and Rommell (2002) suggested probably reflected life history events. Kwan (2002) found that the pregnant females she examined were in extremely good body condition with higher than average mean fat depths compared to other reproductive states (Marsh and Kwan in press). In addition, lactating females had less than average mean fat depths, which is not surprising given the energetic costs of lactation. Therefore, we expected that pregnancies would be indicated by wide GLGs (especially Zone A), reflecting superior body conditions of the animals and that lactation would be indicated by narrow GLGs (especially Zone A and/or a pronounced Zone B), reflecting the poorer body condition of the animals due to the energetic demands of lactation. We observed intervals of inconsistent growth in the dugong tusks we examined, but the patterns were not sufficiently distinct and consistent for tusks to be used as reliable records of the life history of dugongs without accompanying life history information. The tusks of the individuals we examined (Appendix 1) exhibited periods of wide spacing followed by narrow spacing, congruent with periods of pregnancy and lactation for all of these individuals' presumed pregnancies. Pregnancies were determined by Kwan (2002) from the number of scars caused by attachment of the placenta in the uterus. Even though it is not known how advanced a dugong pregnancy has to be to produce a placental scar, scars will be produced by pregnancies that do not go to full term or for a pregnancy that is followed by the death of the lactating calf. Therefore, in some cases superior growth may not be followed by poor growth because the dugong did not suckle the calf for a prolonged period.

The tusks of dugongs that were lactating when they died almost always showed pattern of GLGs consistent with the high energetic demands of lactation. However, this pattern of wide GLGs followed by narrow GLGs was not limited to inferred periods of lactation. It is logical to expect that stress similar to the energetic demands of lactation may be caused by environmental changes such as seagrass diebacks (Marsh and Kwan in press). Florida manatees may not exhibit the same environmental stresses as dugongs as they are generalist feeders (Hartman 1979) with many foraging options. In contrast, dugongs are seagrass community specialists (Marsh et al. 1982), whose fecundity varies with the condition of their seagrass food as discussed below.

The four males examined in this study were all young, but mature animals, three were in full spermatogenesis; Kwan (2002) classified the testes of the fourth animal as resting *sensu* Marsh et al. (1984c). The spacing of GLGs in the tusks of male dugongs was fairly consistent throughout their lives. This pattern is consistent with Kwan's (2002) finding that there was no significant difference in the body condition of male dugongs amongst different reproductive stages. Therefore, we would expect

that male dugongs would show a consistent pattern of growth/fat deposition through time as observed in the spacing of GLGs in the tusks in this study. In addition, male dugong tusks erupt and wear and so their growth is not constrained by the length of the pre-maxilla as it is in female dugongs (Marsh 1980).

We consider that it would be inappropriate to further investigate the spacing in the GLGs of tusks of both male and female dugongs from Torres Strait as a Masters project because these layers do not show a distinct pattern that can unequivocally be attributed to life history events.

4.2 Dugong Fecundity

Despite the large temporal and spatial differences in dugong fecundity measured by the proportion of calves seen during aerial surveys (Table 1), significant consistent relationships between the proportion of calves and all the external covariates were limited to the Northern Great Barrier Reef region. There was a significant negative relationship between the proportion of calves and the wet season rainfall, the wet season rainfall and anomaly and the SOI, each lagged by two years (Figures 8-10) and a significant positive relationship between the proportion of calves and the NINO 3.4 sea surface temperature index lagged by two years (Figure 11). This association was also evident in the analysis which incorporated the subregions in the Northern Great Barrier Reef region (Figures 13-16). The persistence in the relationship across the four external covariates suggests it is real, especially as it is congruent with our knowledge of dugong life history and the dynamic responses of the coastal seagrasses in which dugongs feed to changes in water turbidity and sediment deposition, which in turn are linked to extreme rainfall events (Preen and Marsh 1995, Preen et al. 1995, Longstaff and Dennison 1999, Waycott et al. 2007, Larcombe and Woolfe 1999).

Other significant relationships between the proportion of calves and some external covariates were found for the survey region in Western Australia and the Moreton Bay region. In the Western Australia region there was a significant negative relationship between the proportion of calves and the SOI and a significant positive relationship between the proportion of calves and NINO3.4, each lagged by two years; but the relationship with rainfall or rainfall anomalies could not be tested because of the low variation in rainfall. It seems likely that the seagrass beds in Western Australia were destroyed by the cyclonic waves associated with category 5 Cyclone Vance in 1999 rather than by poor water quality associated with plumes of terrestrial runoff per se. This result is not surprising as the coastal environment in Western Australia is very different from the east coast and six severe tropical cyclones have been reported in recent years. The water is much clearer and the seagrass on which dugongs feed less likely to be occurring close to the limits of light availability and cyclones cause widespread damage (Gales et al. 2004)

In the Moreton Bay region, the proportion of calves positively covaried with rainfall lagged by two years, suggesting that rainfall may have been having a positive effect on the dugong's food supply as claimed by Welsby (1905). In addition there was a negative relationship between the proportion of calves and rainfall and rainfall anomalies each lagged by three years (which may have been a consequence of the two-year positive relationship), but no relationship with SOI or NINO 3.4. Most of the

dugongs in Moreton Bay occur in the eastern bay where conditions are much more oceanic than in any other area which supports large numbers of dugongs on the east coast of Australia and the impact of rainfall in the coastal catchments would be expected to be different.

Nonetheless, we consider that the significant relationships we found external to the Northern Great Barrier Reef region should be regarded as preliminary because they were not consistent across all four external covariates and were based on more limited datasets.

In the Northern Great Barrier Reef region, the negative relationship between the proportion of calves and the rainfall with a two year lag is presumably a result of: (1) the negative impact of increased turbidity on some of the coastal seagrass species eaten by dugongs (Preen et al. 1995, Longstaff and Dennison 1999); (2) the need for dugongs to be in good condition prior to and during pregnancy and lactation (Kwan 2002, Marsh and Kwan in press) and (3) the life history of the dugong: pregnancy lasts approximately 14 months and lactation up to 18 months (Marsh et al. 1984 b, Boyd et al. 1999, Kwan 2002). The significant lag between green turtle fecundity and the SOI is also two years (Limpus and Nicholls 1988, 2000). This lag is attributed to the time required for female green turtles to accumulate the fat reserves necessary for breeding.

As listed below, there are several possible explanations for the fact that these relationships were not significant for dugongs in the other survey regions.

1. As pointed out above, the regional differences in survey years resulted in different distributions of values of the external covariates (total wet season rainfall, wet season anomaly, SOI and NINO3.4). For Western Australia, the range of wet season rainfall values observed over the survey years (or the lags thereof) was too small to test the association between calf proportions and rainfall/rainfall anomalies.
2. At least some dugongs move, apparently in response to changes in their seagrass food (Marsh et al. 1997, 2004a, 2006, 2007; Gales et al. 2004) and may be able to compensate for local seagrass loss to some extent, especially in areas where the dugong population is reduced such as in the Southern Great Barrier Reef (Marsh et al. 2005). Conversely, such movements may cause declines in fecundity as a result of over-grazing in areas which have not been exposed to pulses in turbidity but where dugongs have emigrated because of seagrass loss in other areas. The apparent emigration of dugongs from Hervey Bay to Moreton Bay in the mid 1990s (Preen and Marsh 1995) is a plausible reason for the low calf counts in Moreton Bay in 1995. Emigration from Exmouth Gulf may also explain the low calf counts in Shark Bay in 1999 (Gales et al. 2004).
3. The seasonal indices of weather that we used may not have adequately reflected the local impact of pulsed rainfall events (e.g. it is possible to have days of extreme rainfall which produce pulses of turbid water in a season of average rainfall).
4. Seagrass loss can result from other natural environmental factors such as light deprivation resulting from increases in turbidity resulting from sediment resuspension during prolonged periods of high winds (Saint-Cast2008), an

external covariate not tested here. Saint-Cast (2008) believes that sediment resuspension is the major cause of seagrass dieback in Torres Strait, a region where there is strong evidence to suggest that the seagrass dieback in the mid 1970s caused a major decline in dugong fecundity (Marsh and Kwan 2008).

5. There are species differences in the capacity of seagrass to withstand episodic decreases in light availability (Longstaff and Dennison 1998, Waycott et al. 2007). These differences mean that the negative impact of pulsed turbidity events on seagrass will vary with seagrass community composition. For example, the effect is expected to be less for seagrass communities dominated by later successional species such as *Thalassia* (the major food for dugongs in Torres Strait, Andre *et al.* 2005) than it is for coastal seagrass communities dominated by *Halophila* (Waycott et al. 2007).
6. There are locational differences in the susceptibility of dugong habitats to terrestrial runoff as explained for the region surveyed in Western Australia and for Moreton Bay above.

We consider that it is likely that the relative importance of these factors differed in different survey regions. The absence of a significant result for Hervey Bay is surprising given the impact of the loss of more than 1000 km² of seagrass following two floods and a cyclone in early 1992 (Preen et al. 1995). At least 99 dugongs died and the dugong population of the Bay was reduced largely by emigration from an estimated 1753 \pm se 388 in 1988 to 600 \pm 126 in December 1993 (Preen and Marsh 1995). The proportion of the population that remained that were calves plummeted to 2.2% in December 1993 from 22% in August 1988 and November 1992. This result presumably reflected the energetic trauma associated with the period of starvation following the loss of seagrass. In an extension of this project, we propose to collaborate with meteorologists to reanalyse the calf proportions using some measure of pulsed rainfall events and to analyse records of dugong strandings in the same way.

Nonetheless, the absence of an overall association across all survey regions between SOI and the proportion of calves, indicates that the association between dugong fecundity and the SOI, a very broadscale index of inter-annual variations in weather, is less marked in dugongs than it is for green turtles (Limpus and Nicholls 1998 and 2000). A plausible reason for this difference is that at least some dugongs move in response to seagrass loss (Preen and Marsh 1995, Gales et al. 2004, Marsh et al., 2004a, 2006, 2007) while green turtles show strong site philopatry to a particular foraging area (Musick and Limpus 1997).

The negative impact on dugong fecundity of the loss of coastal seagrass associated with exceptionally high rainfall and other extreme weather events is the major concern about the impact of climate change on dugongs (Waycott et al. 2007, Lawler et al. 2007).

5. Conclusions

- 5.1 The spacing in the GLGs of the tusks of both female and male dugongs from Torres Strait does not show a distinct pattern that can unequivocally be attributed to life history events such as the onset of sexual maturity or calving. Plausible scenarios can be developed to explain the pattern of GLG deposition in individual

dugongs for which life history information is available from carcasses. However, these patterns are too uncertain for the pattern of GLG deposition in the tusk of a dugong from a tropical region such as Torres Strait to be used as a unambiguous record of its life-history.

- 5.2 The temporal and spatial differences in dugong fecundity measured by the proportion of calves seen during aerial surveys are large. There was a significant negative relationship between the proportion of calves and the wet season rainfall, the wet season rainfall and anomaly and the SOI, and a positive relationship between proportion of calves and the wet season NINO 3.4 sea surface temperature index, each lagged by two years in the Northern Great Barrier Region. In the Western Australian survey region there was a significant negative relationship between the proportion of calves and the SOI and a significant positive relationship between the proportion of calves and NINO3.4, each lagged by two years; but the relationship with rainfall or rainfall anomalies could not be tested because of the low variation in rainfall. In the Moreton Bay region, the proportion of calves positively covaried with rainfall lagged by two years and negatively covaried with rainfall and rainfall anomalies each lagged by three years (which may have been a consequence of the two-year positive relationship), but no relationship with SOI or NINO 3.4. The results for Western Australian and Moreton Bay are tentative as they were not robust over all covariates. Similar relationships may exist for other regions and that the data are not yet adequate to test such relationships (Type 2 error).

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8. Tables

Table 1: Dates and locations of the aerial surveys from which the data on dugong calf counts were extracted.

Locality	Date of Survey	Number of calves sighted	Total number of dugongs sighted	Reference
Shark Bay WA	Oct., Dec. 1981, Feb., Apr., June, Aug. 1982	227	2178	Anderson (1986)
	July 1989	68	354	Marsh <i>et al.</i> (1994)
	July 1994	48	290	Preen <i>et al.</i> (1997)
	July 1999	26	534	Gales <i>et al.</i> (2004)
	Feb. 2002	14	386	Holley <i>et al.</i> (2006)
	July 2007	85	398	Hodgson (2007)
Exmouth Gulf-Ningaloo WA	July 1989	14	57	Marsh <i>et al.</i> (1994)
	July 1994	8	40	Preen <i>et al.</i> (1997)
	July 1999	0	29	Gales <i>et al.</i> (2004)
	June 2007	0	35	Hodgson (2007)
Northern Coast NT	Dec. 1983	4	133	Bayliss (1986)
NT coast of Gulf of Carpentaria	Aug. 1984, Feb. 1985	46	435	Bayliss and Freeland (1989)
QLD coast of Gulf of Carpentaria	Apr. 1975	11	247	Ligon (1976)
	Apr. 1977	0	73	Heinsohn (1977)
	Nov. 1978	1	96	Heinsohn and Marsh (1978)
	Dec. 1991	8	123	Database/Marsh and Lawler (1993)
	Nov. 1997	11	128	Marsh <i>et al.</i> (1998)
	Nov. 2006	2	30	Database/Marsh unpublished
QLD and NT coast of Gulf of Carpentaria	Nov. 2007	58	464	Database/Marsh unpublished
Torres Strait	Apr. 1975	11	122	Ligon (1976)
	Nov. 1983	32	223	Marsh (1986)
	Nov. 1987, Mar. 1988	65	472	Marsh and Saalfeld (1988)
	Nov.–Dec. 1991	57	419	Database/Marsh and Lawler (1992)
	Nov. 1996	73	602	Database/Marsh <i>et al.</i> (1997)
	Nov. 2001	29	293	Database/Marsh <i>et al.</i> (2004b)
	Nov. 2006	57	374	Database/Marsh <i>et al.</i> (2007)

Table 1 continued

NGBR region north of Cape Bedford	Nov. 1974	5	171	Heinsohn <i>et al.</i> (1976)
	Nov. 1976	23	209	Heinsohn (1976)
	June, Nov. 1978	2	1175	Heinsohn and Marsh (1978)
	Nov. 1984	11	95	Marsh (1985)
	Nov. 1985	39	309	Marsh and Saalfeld (1987,1989)
	Nov. 1990	64	503	Database/Marsh <i>et al.</i> (1993)
	Nov. 1995	59	493	Database/Marsh and Corkeron (1996)
	Nov. 2000	72	615	Database
	Nov. 2006	38	532	Database/Marsh <i>et al.</i> (2007)
GBR region south of Cape Bedford	Sep.- Dec. 1974	11	161	Heinsohn (1976)
	1975	15	396	Heinsohn (1976)
	Feb., Apr., June, Aug., Oct. 1976	9	175	Heinsohn (1976)
	June 1977	4	98	Heinsohn (1977)
	Jan., July, Aug., Nov. 1978	9	124	Heinsohn and Marsh (1978)
	Aug., Nov. 1979	13	411	Heinsohn and Marsh (1980)
	Sep. 1986	15	144	Marsh and Saalfeld (1990)
	Nov. 1992	13	101	Marsh <i>et al.</i> (1994)
	Nov. 1994	11	102	Marsh <i>et al.</i> (1995)
	Nov. 1999	35	198	Marsh and Lawler (2001)
	Nov. 2005	25	265	Database/Marsh <i>et al.</i> (2006)
Hervey Bay	Aug. 1979	11	156	Heinsohn and Marsh (1980)
	Aug. 1988	44	199	Database/Marsh <i>et al.</i> (1990)
	Nov.1992	19	87	Database/Preen and Marsh (1995)
	Dec. 1993	1	45	Database/Preen and Marsh (1995)
	Nov. 1994	2	130	Marsh <i>et al.</i> (1995)
	Oct-Dec. 1999	17	117	Marsh and Lawler (2001)
	Apr. 2001	8	94	Lawler (2002)
	Nov. 2001	13	156	Lawler (2002)
	Nov. 2005	20	278	Database/Marsh <i>et al.</i> (2006)

Table 1 continued

Moreton Bay	May, Sep. 1976	28	291	Heinsohn 1976
	May 1977	14	210	Heinsohn 1977
	Aug. 1979	17	307	Heinsohn and Marsh (1980)
	Jan., Mar., May, July, Sep., Dec. 1995	104	3485	Lanyon (2003)
	Oct- Dec. 1999	0*	12	Marsh and Lawler (2001)
	Dec. 2000, Apr 2001	41	410	Lawler (2002)
	Nov. 2001	41	330	Lawler (2002)
	Nov. 2005	10	97	Database/Marsh et al. (2006)

*Region not comprehensively surveyed so result may be biased.

Table 2: Date of death and life-history information for the dugongs whose tusks were measured in this study.

ID number	Date of death	Sex	Reproductive status	Number of placental scars	Status of mammary glands	Size of foetus (cm) if present	Status of ovaries/testes
MD19	16/06/1998	Female	UPG	0		57.2	
MD41	9/07/1998	Female	U/PFO	4			Follicular
MD56	7/08/1998	Female	POV	2			Ovulating
MD58	9/08/1998	Female	ULC	1	Lactating		
MD60	21/08/1998	Female	PPG	2		105.2	
MD73	30/09/1998	Female	ULC	1	Lactating		
MD96	6/11/1998	Female	PLC	4	Lactating		
MD98	14/03/1999	Female	PLC	4	Lactating		
MD101	19/03/1999	Female	ULC	1	Lactating		
MD108	1/04/1999	Female	PPG	1		30.08	
MD122	20/04/1999	Female	ULC	1	Lactating		
MD124	21/04/1999	Female	PLC	4	Lactating		
MD138	18/05/1999	Female	PLC	2	Lactating		
MD148	16/07/1999	Female	PLC	3	Lactating		
MD155	20/07/1999	Female	ULC	1	Lactating		
MD161	26/05/1999	Female	PLC	3	Lactating		
MD21	21/04/1999	Male	FSP				Active
MD28	5/07/1998	Male	FSP				Active
MD123	21/04/1999	Male	RST				Resting
MD26	5/07/1998	Male	FSP				Active

UPG = primigravid pregnant; PPG = multiparous pregnant; ULC = uniparous lactating; PLC = multiparous lactating; POV = multiparous ovulating; FSP = fully spermatogenic; RST = resting mature male.

Table 3: Methods investigated to highlight and measure GLGs in dugong tusks and found unsuitable.

Details of Methodology	Comments
<i>Longitudinal thin sections</i>	
1 mm section of a longitudinal section of a 12.5cm long tusk was cut with a 20cm diamond-edged blade. The sections was ground to 500um on a grinding wheel and then polished on both sides using progressively finer grades of wet and dry sandpaper.	GLGs were not evident when viewed through a microscope using transmitted light.
500µm section was viewed with fluorescent light under a microscope	The GLGs did not fluoresce.
500µm section was stained in Mayers Haematocilin and Mayers Methylene blue	GLGs were not evident and the stain made the section curl up.
A 500µm section was acid-etched and stained	The etching caused the tusk section to split; the stain caused it to curl..
100µm longitudinal thin section was prepared by cutting a 1mm section, polishing one side on a grinding wheel, sticking the polished side to a glass slide and then polishing the other side.	The thin section broke apart on the slide and GLGs were not evident using the naked eye or a microscope.
Both the 100µm and the 500µm sections were viewed under UV light.	The GLGs did not fluoresce.
Previous studies on cetaceans have made longitudinal sections by cutting the tooth into pieces and making a montage.	Such an approach is appropriate for age determination, but is not appropriate for measuring GLG widths because critical information is lost where the cuts are made.
<i>Transilluminating scanner (AIMS)</i>	
A 1mm thin section was scanned	The sharpness of the images was not as good as that of the digital photographs.

Table 3 continued.

<i>x-ray (Mater Hospital)</i>	
Various different thicknesses of sections were trialled	1mm section worked the best
Various settings were trialled on different x-ray machines	<p>The mammography machine worked best because it is designed to detect fine-scale features.</p> <p>25kv, 9 MAs worked, but only showed lines up on one side.</p> <p>We were not able to use the mammography machine because our long exposure times wear out the tube too quickly.</p> <p>Trialling other machines would have required much more trial and error to get the setting right, but may not have resulted in adequate images.</p> <p>x-ray is expensive (\$300 per hour) and therefore would not be a very useful technique for processing many samples as would be required if tusks could be used as a proxy for determining life history characteristics.</p>
<i>Scanning Electron Microscopy (AAC – JCU)</i>	
Tried an acid etched-half tusk and 1mm thin section	It was not possible to create a vacuum in the chamber to coat the tusks in carbon, which is needed to use SEM
<i>Backscatter SEM</i>	
Investigated the possibility of using this technique	Requires very expensive equipment for specimens the size of a tusk, which is not available in Townsville.
<i>Micro-CT</i>	
Adelaide Microscopy, Adelaide University	This technique was unsuccessful. It might be possible to achieve an image with thin sections. This technique is very time-consuming taking about 7 hours to scan each sample.

Table 4 Weather stations from which rainfall totals and anomalies were extracted.

Region	Data available	Weather stations	Assumptions for pooling
Western Australia			
Shark Bay	Oct., Dec., 1981, Feb., Apr., June, Aug. 1982	Denham; Learmonth AP; Carnarvon	Gales <i>et al.</i> (2004) indicated that it is likely that movement of animals from Exmouth affected the situation in Shark Bay; Lag years 1976/77 – 1980/81
	July 1989	Denham; Learmonth AP; Carnarvon	Lag years 1984/85 – 1988/89.
	July 1994		Lag years 1989/90 – 1993/94
	July 1999		Lag years 1994/95 – 1998/99
	Feb. 2002		Lag years 1996/97 – 2000/01
	June 2007		Lag years 2002/03 – 2006/07
Ningaloo/ Exmouth	July 1989	Denham; Learmonth AP; Carnarvon	Gales <i>et al.</i> (2004) indicated that it is likely that movement of animals from Exmouth affected the situation in Shark Bay; Lag years 1984/85 – 1988/89.
	July 1994		Lag years 1989/90 – 1993/94
	July 1999		Lag years 1994/95 – 1998/99
	June 2007		Lag years 2002/03 – 2006/07
Northern Territory			
Top end	Nov. 1983	Darwin; Waruwi; Black Point	Survey was done between Daly River and Mililingimbi. These weather stations are the only ones in that region with rainfall data for the NWS months for the lag years preceding the survey (1978/79-1982/83).

Table 4 continued

Gulf of Carpentaria			
Western Gulf	Aug. 1984/Feb. 1985	Angurugu; Centre Island	Survey was done between south of Yirrkala and south of Borrooloola. These weather stations are the only ones in that region with rainfall data for the NWS months for the lag years preceding the survey (1979/1980 -1983/1984)
Eastern Gulf		Weipa (Eastern Ave); Karumba Airport/Normanton; Mornington Is.	Surveys were done between Cape York and Bayley Point. All of these weather stations were used for all surveys even if the survey only covered part of the region because it is likely that dugongs move among these places through time. These weather stations are the only ones in the region of the surveys with rainfall data for the NWS months for the lag years preceding the surveys (see below)
Staaten River – Weipa	Apr. 1975		Lag years 1969/70- 1973/74
Karumba – Bayley Pt.	Apr. 1977		Lag years 1971/72 – 1975/76.
Cape York – Karumba	Nov. 1978		Lag years 1973/74 – 1977/78.
Karumba – Bayley Pt.	Dec. 1991		Lag years 1986/87 – 1990/91.
Eastern Gulf	Nov. 1997		Lag years 1992/93 – 1996/97
Cape York – Weipa	Nov. 2006		Lag years 2001/02 – 2005/2006
Whole Gulf	Nov. 2007	Alyangula; Centre Island; Weipa (Eastern Ave); Karumba Airport/Normanton; Mornington Is.	The survey covered the Northern Territory and Queensland sides of the Gulf of Carpentaria. These weather stations are the only ones in the region of the survey with rainfall data for the NWS months for the lag years preceding the survey (2002/2003 – 2006/2007) and were used in the previous surveys. Note: Alyangula was used instead of Angurugu because data were not available for Angurugu at this time or for Alyangula for the previous surveys.

Table 4 continued

Torres Strait			
		Thursday Island Township/ Horn Island Airport (Thursday Is. MO – 30-year average)	These weather stations are the only ones in the survey region with rainfall data for the NWS months for the lag years preceding the surveys. Thursday Island Township and Horn Island had data for different years. Both had data for the 2005/06 NWS year so the totals from both weather stations were averaged for this year.
	Apr. 1975	Thursday Island Township	Lag years 1969/70 – 1973/74
	Nov. 1983	Thursday Island Township	Lag years 1978/79 – 1982/83
	Nov. 1987, Mar. 1988	Thursday Island Township	Lag years 1982/83 – 1986/87
	Nov. 1991	Thursday Island Township	Lag years 1986/97 – 1990/91
	Nov. 1996	Thursday Island Township/Horn Island Airport	Lag years 1991/92 – 1994/95(TI); 1995(TI)/1996(HI)
	Nov. 2001	Horn Island Airport	Lag years 1996/97 – 2000/01
	Nov.2006	Thursday Island Township/Horn Island Airport	Lag years 2001/02 – 2004/05(HI); 2005/06 (TI and HI).
Northern GBR			
(Blocks 1-5)		Cooktown PO/Cooktown;	These weather station are the only one in the survey region with rainfall data for the NWS months for the lag years preceding the surveys; Different weather stations were used for Blocks 1-5 and Blocks 6- 15 because of the large distances between these areas.
Blocks 1-5 (Cooktown to Princess Charlotte Bay)	Nov. 1974	Cooktown PO	Lag years 1969/70 – 1973/74

Table 4 continued

Blocks 2 and 4 (Lookout Point to Starcke River)	Nov. 1976	Cooktown PO	Lag years 1971/72-1975/76
Blocks 1-5 (Cooktown to Princess Charlotte Bay)	Jun, Nov. 1978	Cooktown PO	Lag years 1973/74-1977/78
Blocks 1-4 (Cooktown to Cape Melville)	Nov. 1984	Cooktown PO	Lag years 1979/80-1983/84
Blocks 1-4; 5 (Cooktown to Cape Melville; Princess Charlotte Bay)	Nov. 1985	Cooktown PO	Lag years 1980/81-1984/85
Blocks 1-4; 5 (Cooktown to Cape Melville; Princess Charlotte Bay)	Nov. 1990	Cooktown PO/Cooktown	Lag years 1985/86-1989/90
Blocks 1-4; 5 (Cooktown to Cape Melville; Princess Charlotte Bay)	Nov. 1995	Cooktown	Lag years 1990/91-1994/95
Blocks 1-4; 5 (Cooktown to Cape Melville; Princess Charlotte Bay)	Nov. 2000	Cooktown	Lag years 1995/96-1999/2000
Blocks 1-4; 5 (Cooktown to Cape Melville; Princess Charlotte Bay)	Nov. 2006	Cooktown	Lag years 2001/2002-2005/2006

Table 4 continued

(Blocks 6-15)		Lockhart River	This weather station are the only one in the survey region with rainfall data for the NWS months for the lag years preceding the surveys; Different weather stations were used for Blocks 1-5 and Blocks 6-15 because of the large distances between these areas.
Blocks 6-15	Nov. 1978		Lag years 1973/74-1977/78
Blocks 6-14	Nov. 1985		Lag years 1980/81-1984/85
Blocks 6-15	Nov. 1990		Lag years 1985/86-1989/90
Blocks 6-15	Nov. 1995		Lag years 1990/91-1994/95
Blocks 6-15	Nov. 2000		Lag years 1995/96-1999/2000
Blocks 6-15	Nov. 2006		Lag years 2001/2002-2005/2006
Southern GBR			
Central SGBR		Cardwell, Townsville, Ayr, Burdekin Shire Council	Most dugongs in the central SGBR are found near Hinchbrook, Townsville and Upstart bay. These weather stations are near those areas and have rainfall data for the lag years preceding each survey.
C8-10 (Townsville – Hinchbrook)	Sep., Oct., Nov., Dec 1974		Lag years 1969/70-1973/74
C8-10 (Townsville – Hinchbrook)	All 1975		Lag years 1970/71 – 1974/75
C8-10 (Townsville – Hinchbrook)	Feb, Apr, Jun, Aug, Oct., 1976		Lag years 1971/72 – 1975/76
C7&8 Cleveland Bay and Bowling Green Bay	June 1977		Lag years 1972/73 – 1976/77

Table 4 continued

C1-10 (Proserpine – Hinchbrook)	Jan., July, Aug., Nov. 1978		Lag years 1973/74 – 1977/78
C8-10 (Townsville – Hinchbrook)	Nov. 1979		Lag years 1974/75 – 1978/79
C8-11 (Townsville – Hinchbrook)	Sep. 1986		Lag years 1981/82 – 1985/86
C1-12 (Proserpine – Cooktown)	Nov. 1992		Lag years 1987/88 – 1991/92
C1-11 (Proserpine–Dunk Island)	Nov. 1994		Lag years 1989/90 – 1993/94
C1-12 (Proserpine–Cooktown)	Nov. 1999		Lag years 1994/95 – 1998/99
C1-12 (Proserpine–Cooktown)	Nov. 2005		Lag years 2000/01 – 2004/05
Southern SGBR		Pacific Heights; Rockhampton; St Lawrence	Most dugongs in the southern SGBR are in Shoalwater Bay. These weather stations are near Shoalwater Bay and have rainfall data for the NWS months in the lag years preceding the surveys.
S4-5	June, 1975		Lag years 1970/71 – 1974/75
S6-8 (St Lawrence – Proserpine)	July 1978		Lag years 1973/74 – 1977/78
S1-5 (Gladstone – Shoalwater Bay)	Aug., Nov. 1979		Lag years 1974/75 – 1978/79
S1-8 (Bundaberg – Proserpine)	Sep. 1986		Lag years 1981/82 – 1985/86
S1-8 (Bundaberg – Proserpine)	Sep. 1992		Lag years 1987/88 – 1991/92
S1-8 (Bundaberg – Proserpine)	Nov. 1994		Lag years 1989/90 – 1993/94

Table 4 continued

S1-8 (Bundaberg – Proserpine)	Nov. 1999		Lag years 1994/95 – 1998/99
S1-8 (Bundaberg – Proserpine)	Nov. 2005		Lag years 2000/01 – 2004/05
Hervey Bay		Bundaberg Airport Sandy Cape, Maryborough	
	Aug. 1979		Lag years 1974/75 – 1978/79
	July/Aug. 1988		Lag years 1983/84 – 1987/88
	Nov. 1992		Lag years 1987/88- 1991/92
	Dec. 1993		Lag years 1988/89 – 1992/93
	Nov. 1994		Lag years 1989/90 – 1993/94
	Nov. 1999		Lag years 1994/1995 – 1998/99
	Apr. 2001		Lag years 1995/96 – 1999/00
	Nov. 2001		Lag years 1996/97 – 2000/01
	Nov. 2005		Lag years 2000/01 – 2004/05
Moreton Bay		Somerset Dam	Rainfall at Somerset Dam is more likely to affect Moreton Bay than rainfall at Brisbane or Cape Moreton (which are coastal).
	May, Sep. 1976		Lag years 1971/72 – 1975/76
	May 1977		Lag years 1972/73 – 1976/77
	Aug. 1979		Lag years 1974/75 – 1978/79
	Jan, Mar., May, July, Sep., Dec. 1995		Lag years 1990-1991-1994/1995
	Nov. 1999		Lag years 1994/1995 – 1998/99

Table 4 continued

	Nov. 2000, Apr. 2001		Lag years 1995/96 – 1999/00
	Nov. 2001		Lag years 1996/97 – 2000/01
	Nov. 2005		Lag years 2000/01 – 2004/05

Table 5. Reliability of the measurements of the widths of GLGs for each observer.

Tusk	Observer 1				Observer 2			
	Ridge	F	Groove	F	Ridge	F	Groove	F
MD56	-22.9 ¹ %	0.07	-21.4%	0.12	-18.7%	0.21	11.6%	1.65
MD21	-14.6%	0.36	-3.4%	0.84	-16.8%	0.28	7.88%	1.43
MD41	-22.4%	0.08	-23.6%	0.04	-19.4%	0.19	-12.9%	0.43
MD161	-24.5%	0.02	-18.5%	0.22	-14.4%	0.37	-12.5%	0.45

¹Error rates are negative when the F-ratio < 1 because the within observer variance is greater than the among observer variance. This situation was common because different GLGs along a dugong tusk vary in width and therefore it would be expected that the variance in widths among GLGs in the same tusk would be relatively large compared with the repeated measurements of widths of the same GLG among times.

Table 6. Repeatability of the measurements of the widths of the GLGs between observers.

Tusk	Ridge	F	Groove	F
MD56	-24 ¹ .8%	0.006	-20.5%	0.15
MD21	-22.6%	0.08	9.26%	1.51
MD41	-16.4%	0.30	-21.9%	0.10
MD161	22.0%	2.41	-19.3%	0.19

¹Error rates are negative when the F-ratio < 1 because the within observer variance is greater than the among observer variance. This situation was common because different GLGs along a dugong tusk vary in width and therefore it would be expected that the variance in widths among GLGs in the same tusk would be relatively large compared with the repeated measurements of widths of the same GLG among times.

Table 7: The average proportion of calves over all years in each survey region.

Survey Region	Mean proportion calves (95% Confidence Interval)
Western Australia (Shark Bay-Ningaloo)	0.114 (0.086-0.150)
Gulf of Carpentaria	0.086 (0.050-0.144)
Torres Strait	0.129 (0.092-0.180)
Northern Great Barrier Reef	0.076 (0.053-0.108)
Southern Great Barrier Reef	0.074 (0.044-0.119)
Hervey Bay	0.107 (0.062-0.178)
Moreton Bay	0.050 (0.033-0.074)

Table 8: The average proportion of calves over all years in each survey region on the eastern coast of Queensland which was large enough to be divided into sub-regions.

Year	Proportion calves (95% Confidence Interval)
Northern Great Barrier Reef	
1978	0.002 (0.000-0.026)
1984	0.129 (0.041-0.340)
1985	0.128 (0.069-0.224)
1990	0.127 (0.078-0.201)
1995	0.119 (0.071-0.193)
2000	0.121 (0.075-0.188)
2006	0.074 (0.039-0.134)
Southern Great Barrier Reef	
1974	0.075 (0.023-0.213)
1975	0.041 (0.015-0.107)
1976	0.056 (0.016-0.182)
1977	0.045 (0.007-0.246)
1978	0.078 (0.022-0.243)
1979	0.032 (0.011-0.090)
1986	0.107 (0.040-0.257)
1992	0.128 (NA) ^a
1994	0.107 (NA) ^a
1999	0.188 (0.099-0.328)
2005	0.098 (0.045-0.198)
Torres Strait	
1991	0.140 (0.084-0.225)
1996	0.125 (0.079-0.193)
2001	0.100 (0.049-0.193)
2006	0.156 (0.094-0.247)

^aCI not calculated because data for individual blocks was not available.

9. Figures

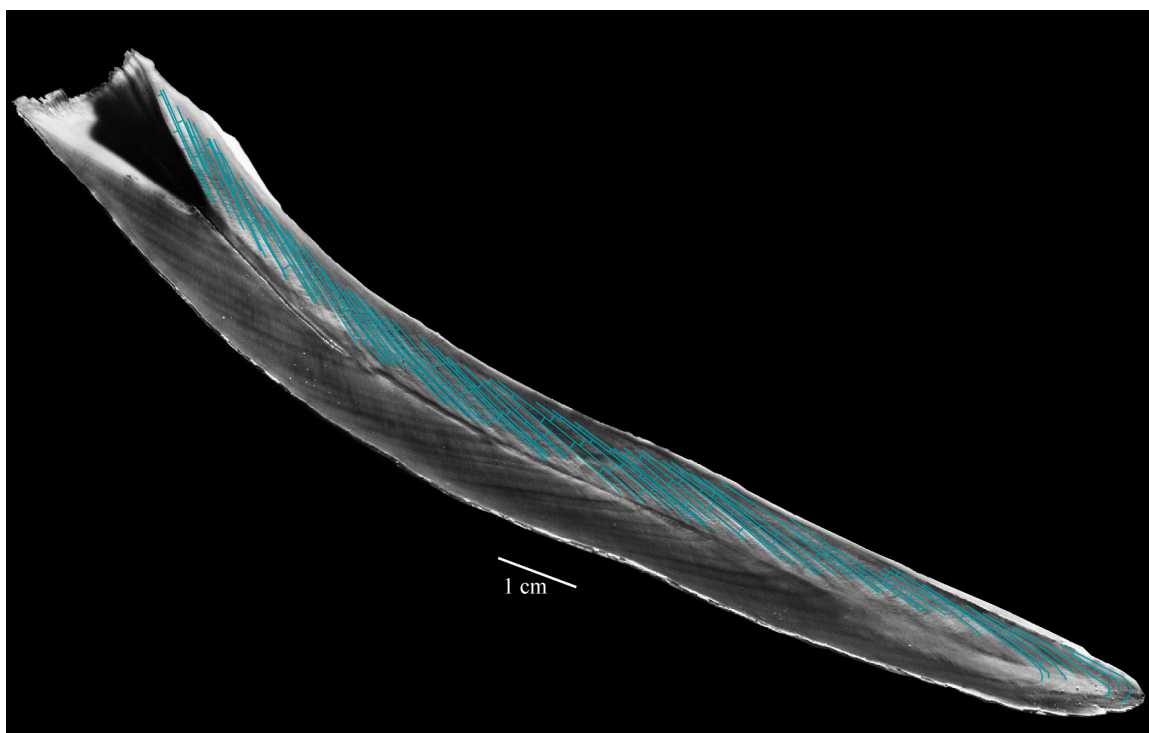


Figure 1. Acid-etched half-tusk of female dugong number MD161 showing growth layer groups labelled for measurement with ArcGIS

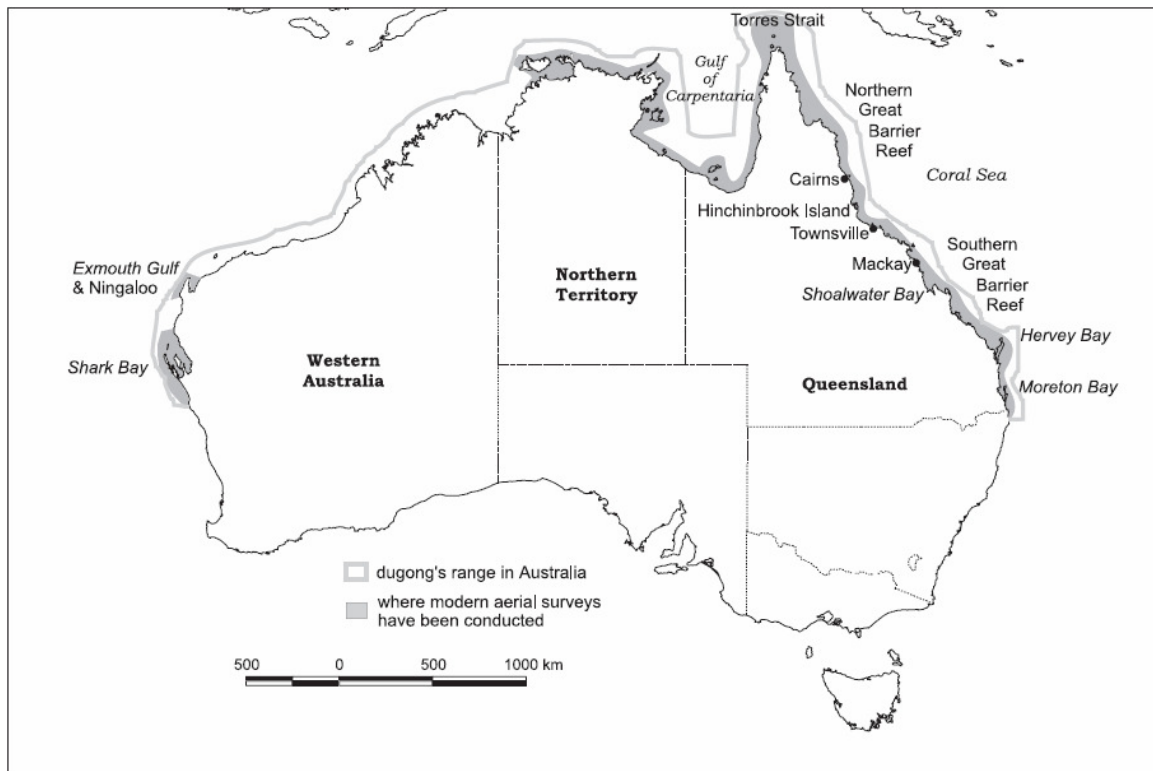


Figure 2. Map of the range of the dugong in Australia, illustrating place names mentioned in Table 1. Adapted from Marsh *et al.* (1999)

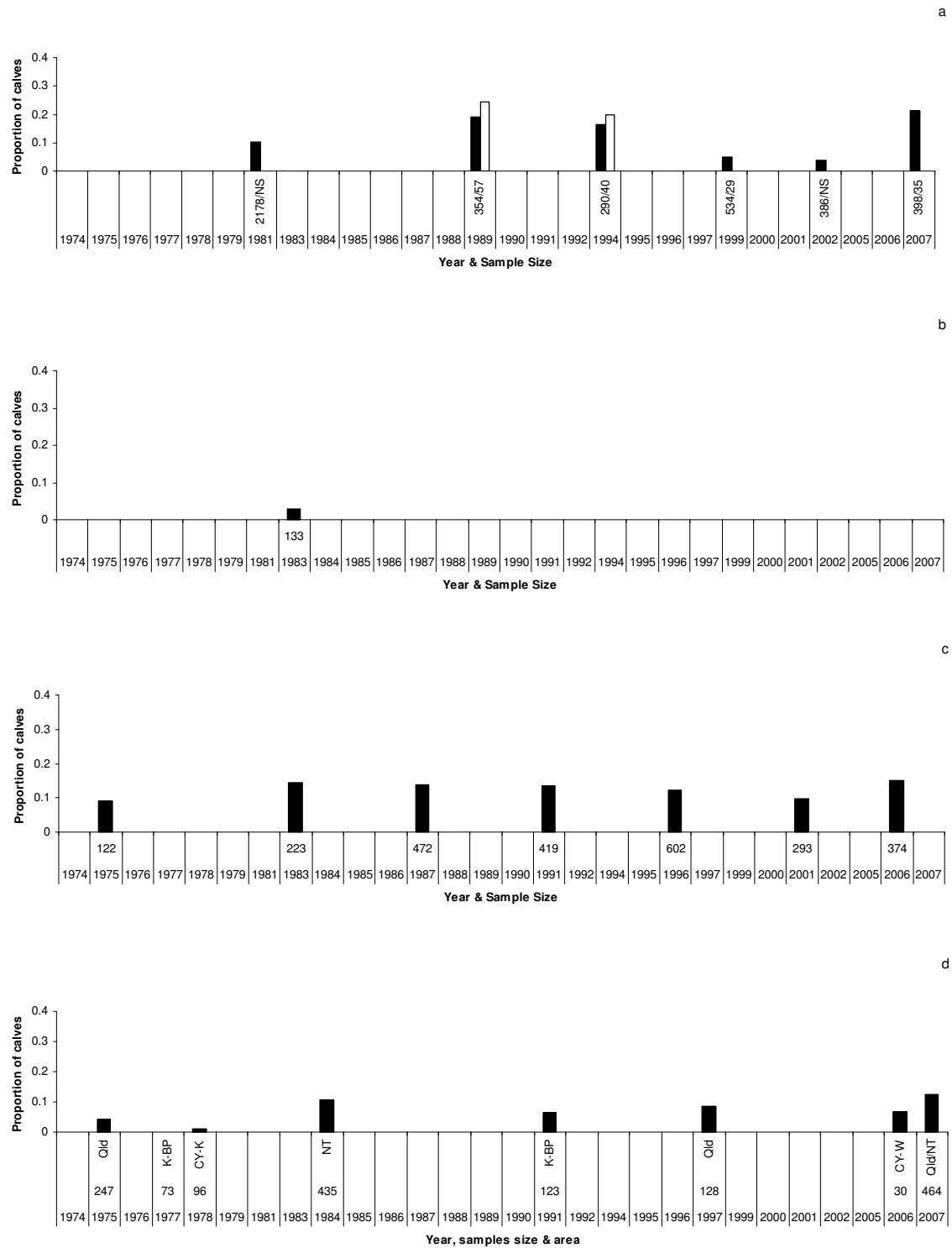
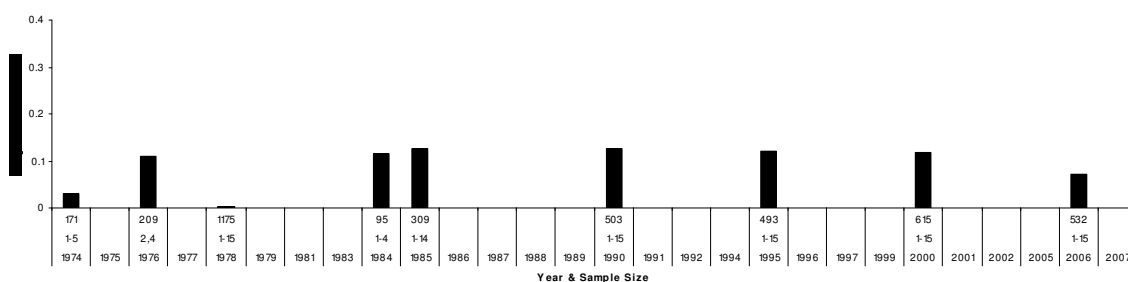
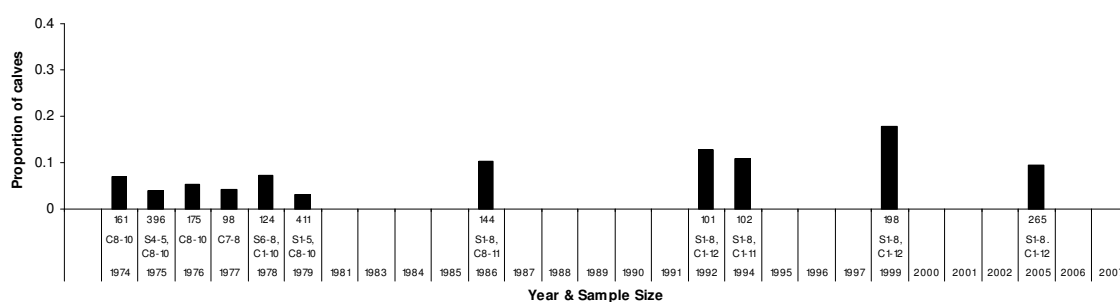


Figure 3. Proportion of calves in the population from aerial survey data from a) Shark Bay (Black bars) and Ningaloo (white bars), Western Australia; b) Northern coast of the Northern Territory; c) Torres Strait; d) Gulf of Carpentaria (K-BP: Karumba - Bayley Point, CY-K: Cape York – Karumba, CY-W: Cape York – Weipa, NT: Northern Territory coast);

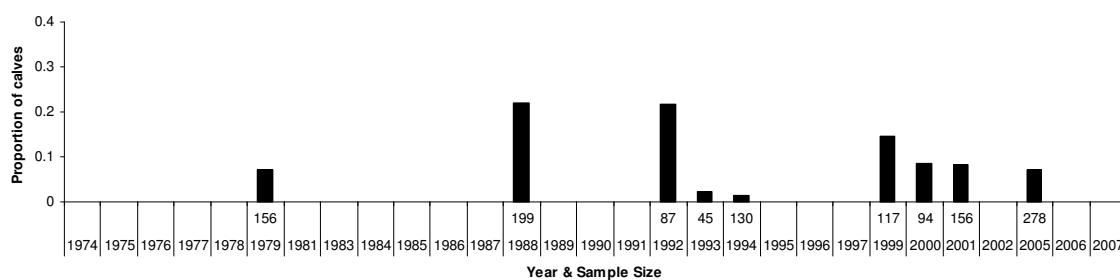
e



f



g



h

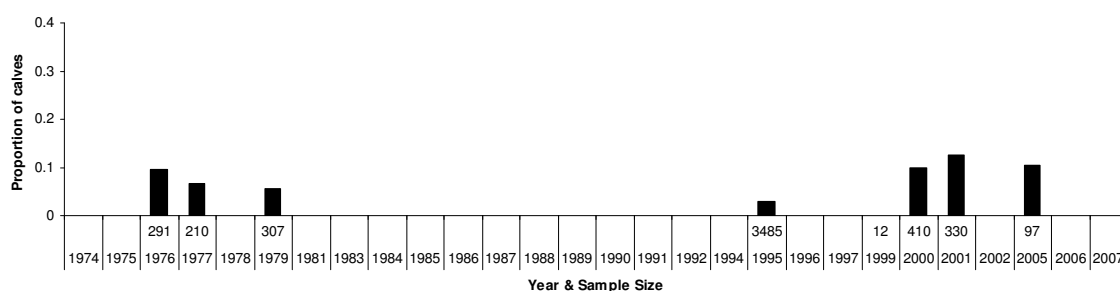


Figure 3 (continued). Proportion of calves in the population from aerial survey data e) Northern Great Barrier Reef; f) Southern Great Barrier Reef; g) Hervey Bay; h) Moreton Bay (Moreton Bay 1999 not comprehensively surveyed so may be biased).

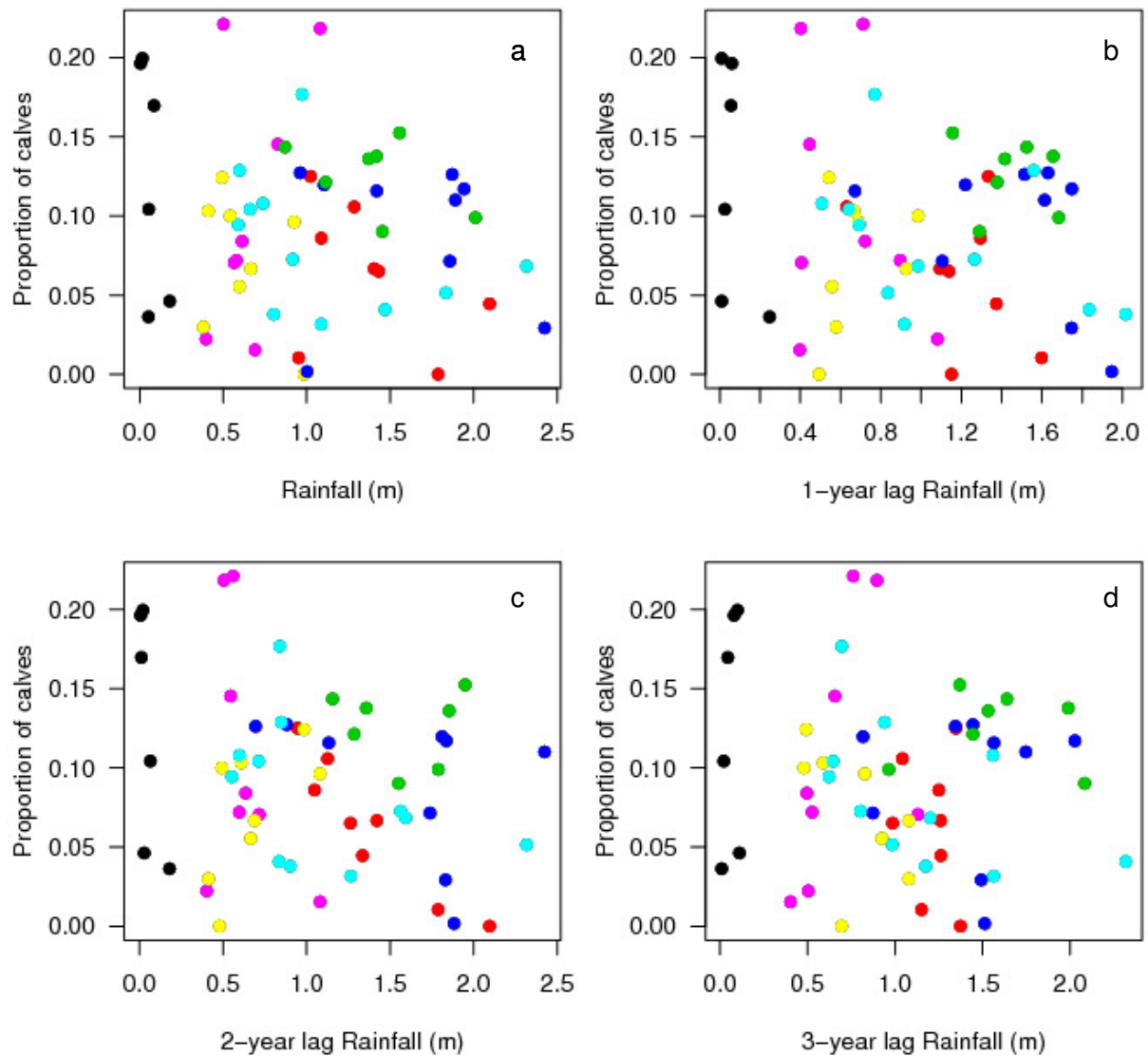


Figure 4. Plots of the raw relationships between the proportion of calves and (a) total wet season rainfall, (b) 1-year lag, (c) 2-year lag and (d) 3-year lag for each survey region. Points are coloured according to the their location as follows: Black, Western Australia; red, Gulf of Carpentaria; green, Torres Strait; dark blue, Northern Great Barrier Reef; light blue, Southern Great Barrier Reef; pink, Hervey Bay; and yellow, Moreton Bay.

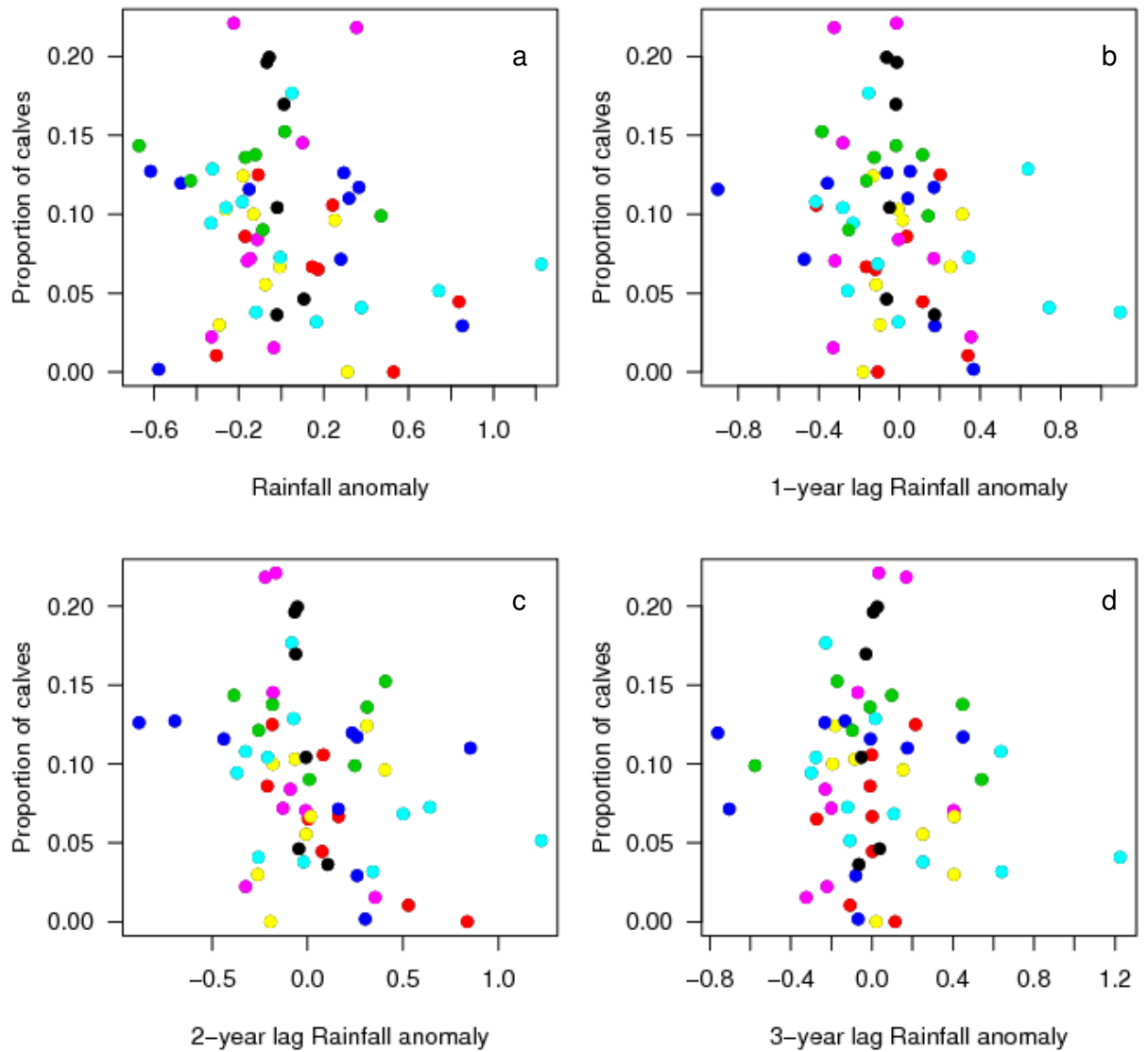


Figure 5. . Plots of the raw relationships between the proportion of calves and (a) wet season rainfall anomalies, (b) 1-year lag, (c) 2-year lag and (d) 3-year lag for each survey region. Points are coloured according to the their location as follows: Black, Western Australia; red, Gulf of Carpentaria; green, Torres Strait; dark blue, Northern Great Barrier Reef; light blue, Southern Great Barrier Reef; pink, Hervey Bay; and yellow, Moreton Bay.

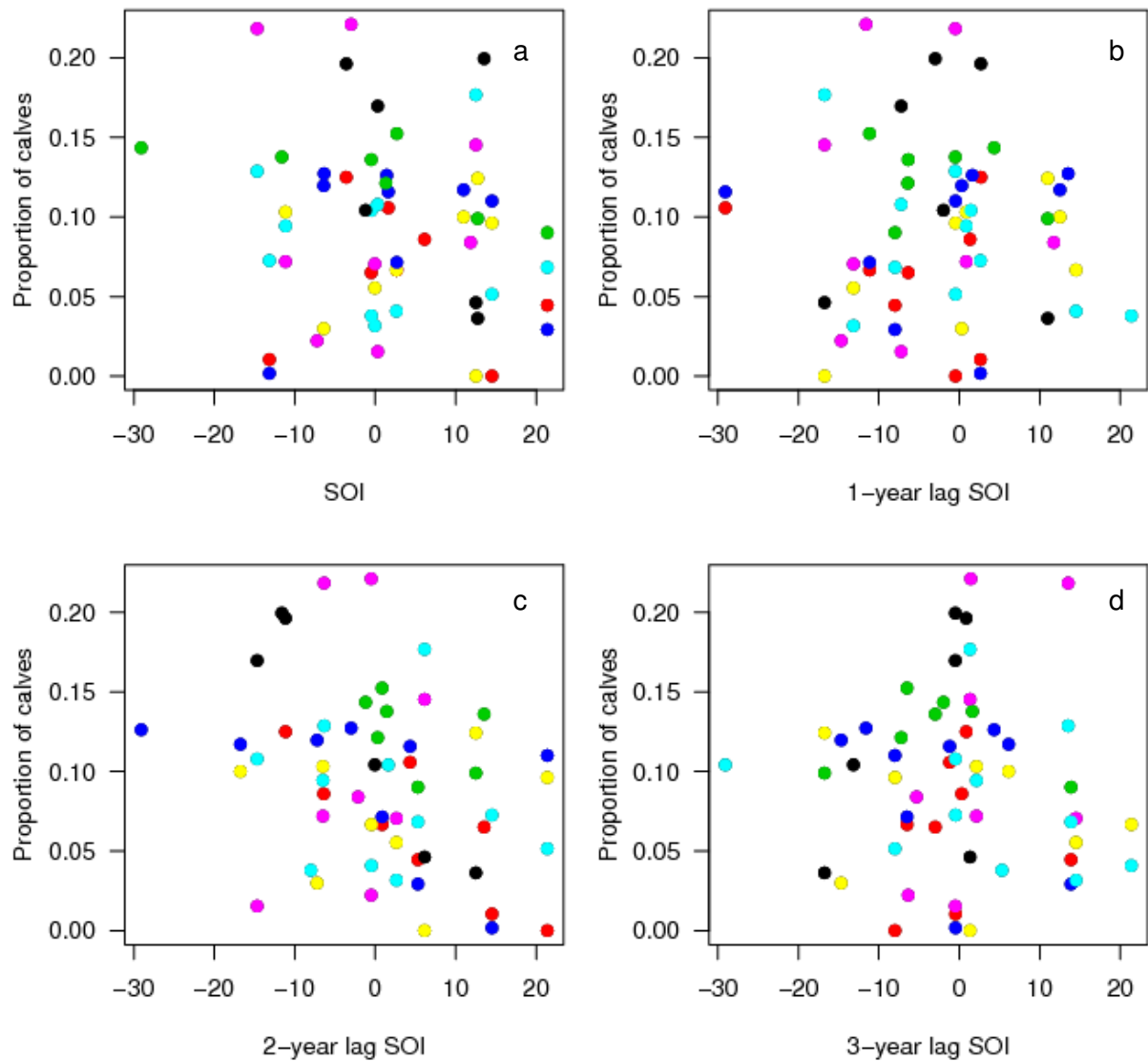


Figure 6. Plots of the raw relationships between the proportion of calves and (a) southern Oscillation Index (SOI) (b) 1-year lag, (c) 2-year lag and (d) 3-year lag for each survey region. Points are coloured according to the their location as follows: Black, Western Australia; red, Gulf of Carpentaria; green, Torres Strait; dark blue, Northern Great Barrier Reef; light blue, Southern Great Barrier Reef; pink, Hervey Bay; and yellow, Moreton Bay.

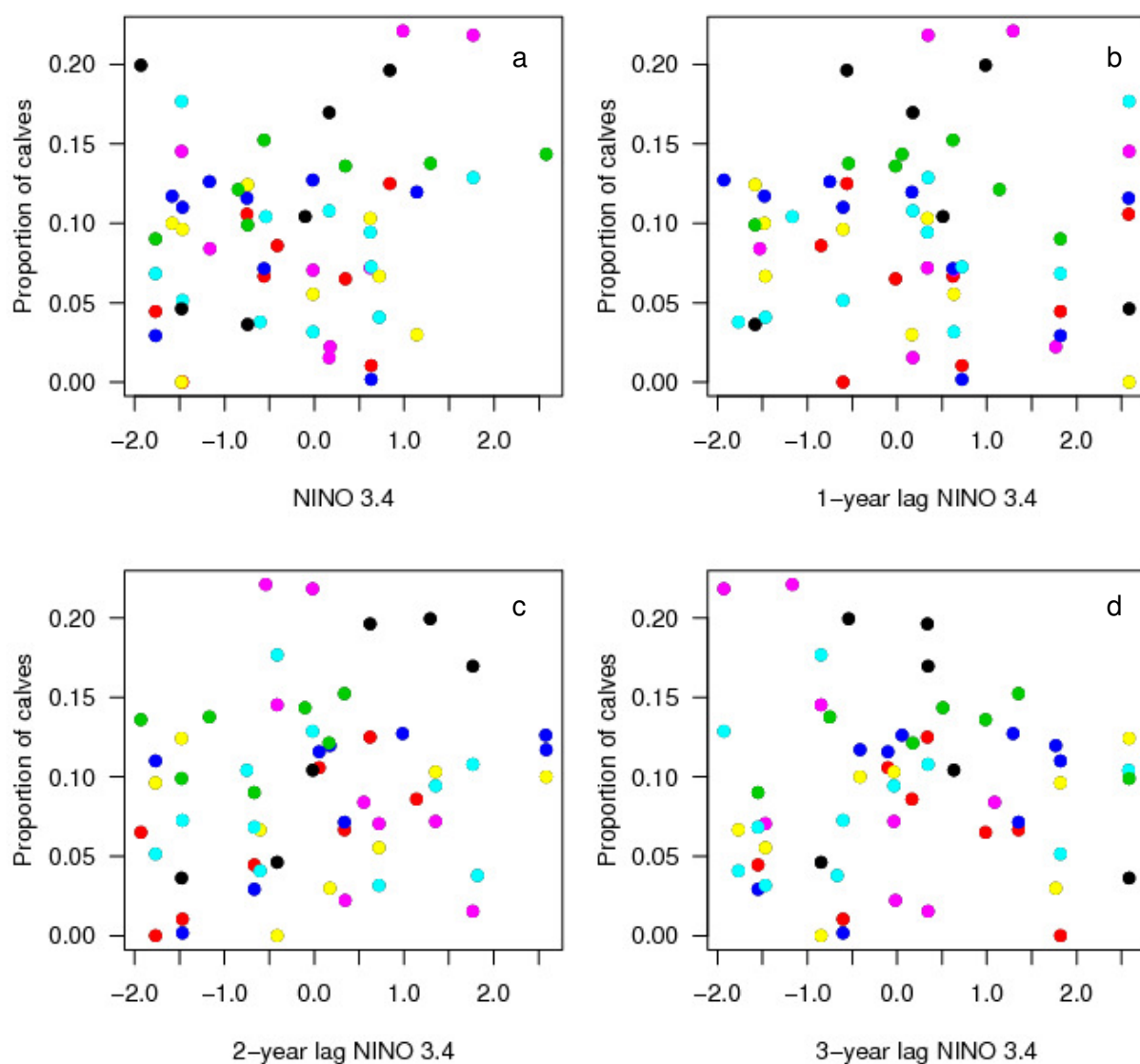


Figure 7. Plots of the raw relationships between the proportion of calves and (a) NINO 3.4 index of sea surface temperature, (b) 1-year lag, (c) 2-year lag and (d) 3-year lag for each survey region. Points are coloured according to the their location as follows: Black, Western Australia; red, Gulf of Carpentaria; green, Torres Strait; dark blue, Northern Great Barrier Reef; light blue, Southern Great Barrier Reef; pink, Hervey Bay; and yellow, Moreton Bay.

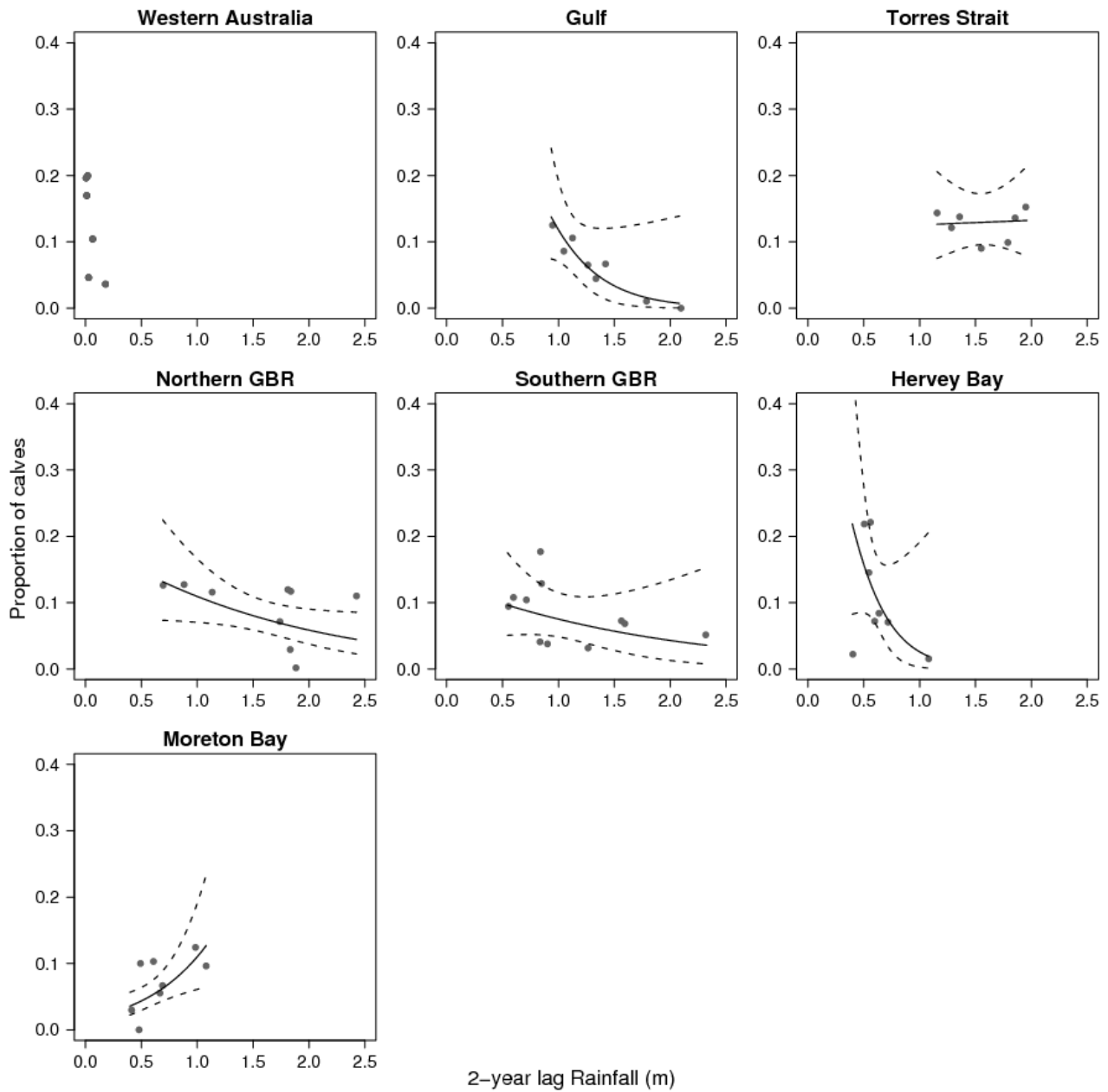


Figure 8. Plots of the estimated relationships between the proportion of dugong calves with total wet season rainfall two years preceding the survey year in each location. Solid lines are estimated mean relationship; dashed lines are 95% confidence intervals.

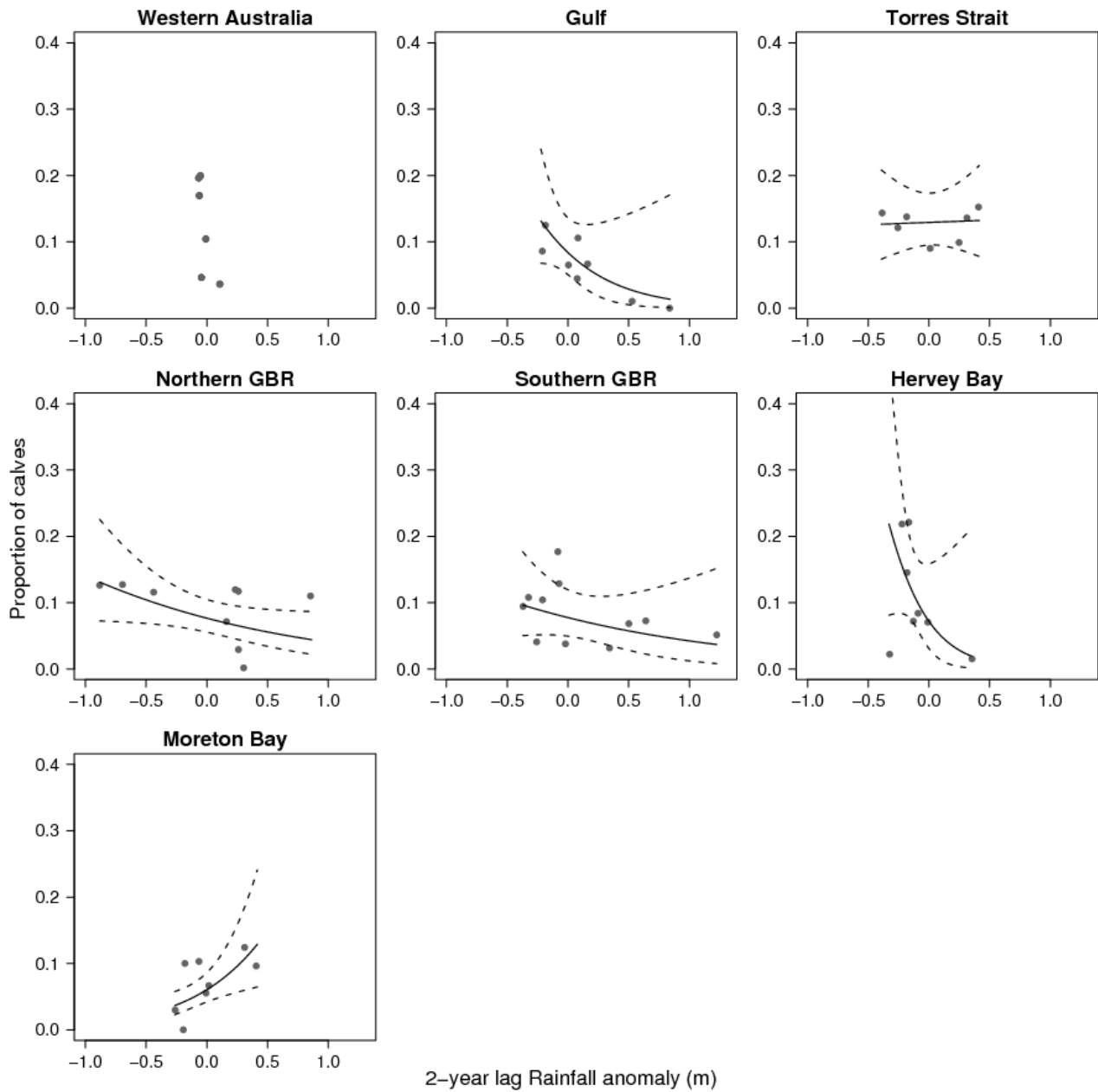


Figure 9. Plots of the estimated relationships between the proportion of dugong calves with wet season rainfall anomalies two years preceding the survey year in each location. Solid lines are estimated mean relationship; dashed lines are 95% confidence intervals.

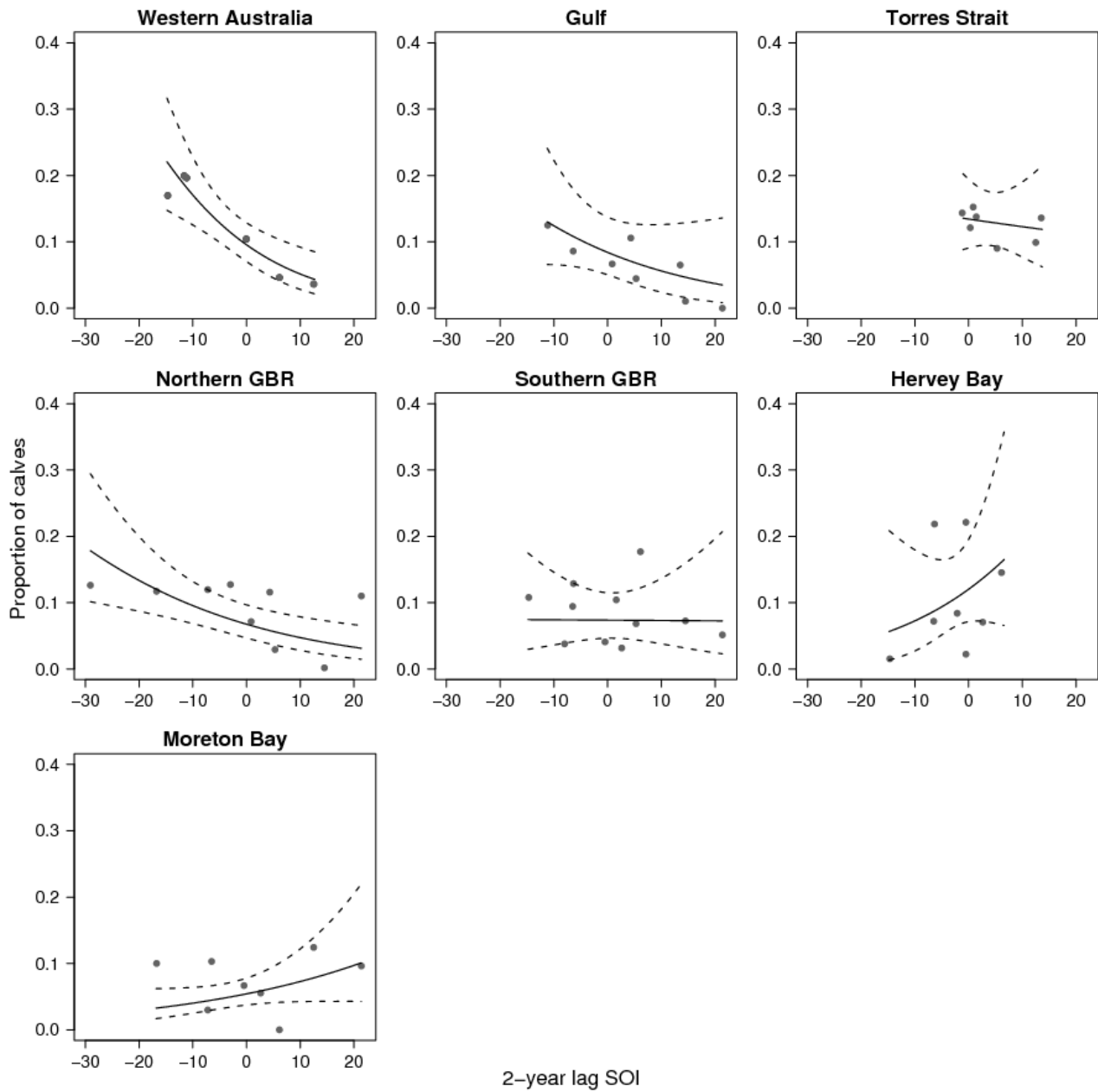


Figure 10. Plots of the estimated relationships between the proportion of dugong calves with the Southern Oscillation Index (SOI) two years preceding the survey year in each location. Solid lines are estimated mean relationship; dashed lines are 95% confidence intervals.

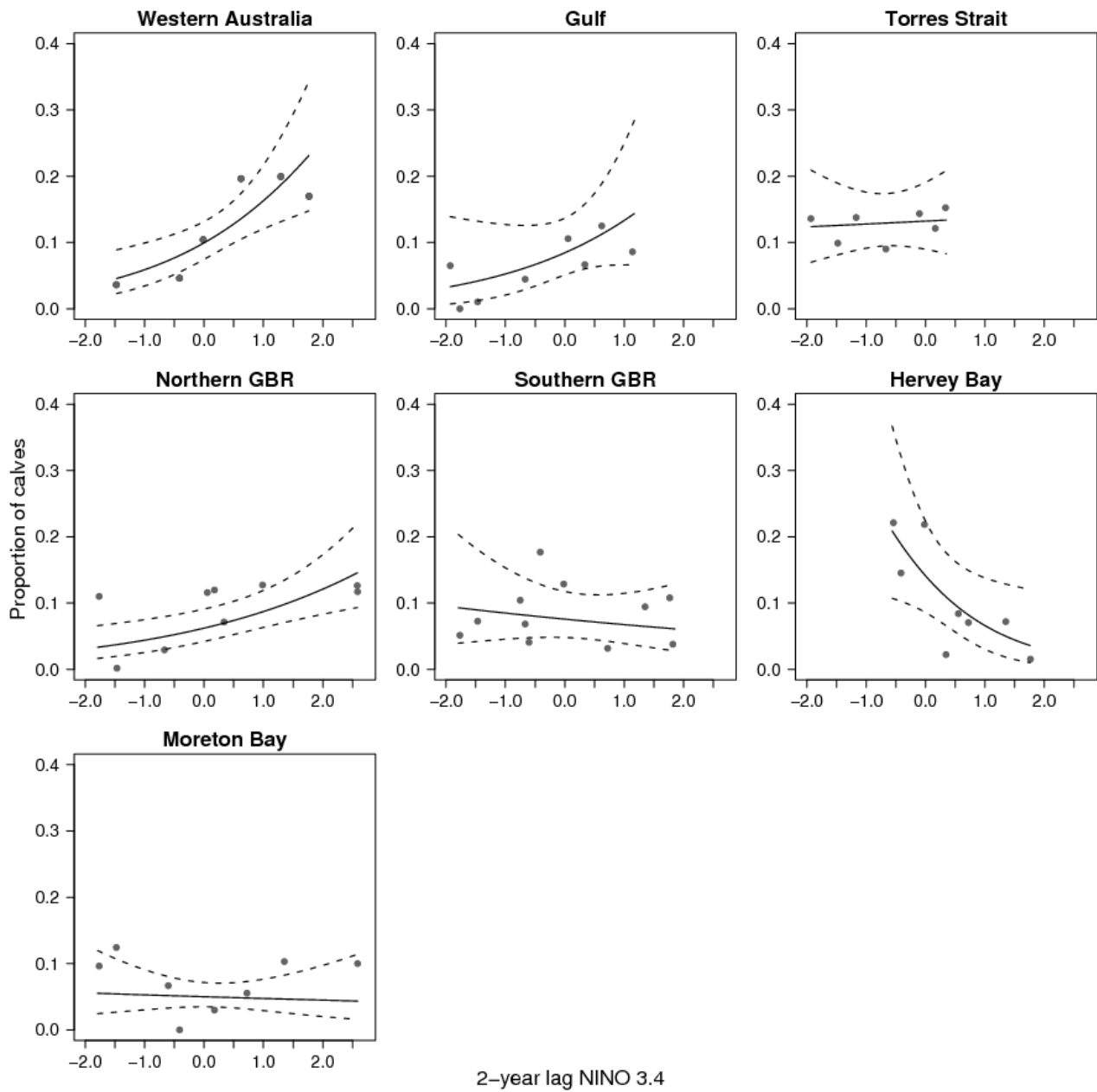


Figure 11. Plots of the estimated relationships between the proportion of dugong calves with the NINO 3.4 index of sea surface temperature two years preceding the survey year in each location. Solid lines are estimated mean relationship; dashed lines are 95% confidence intervals.

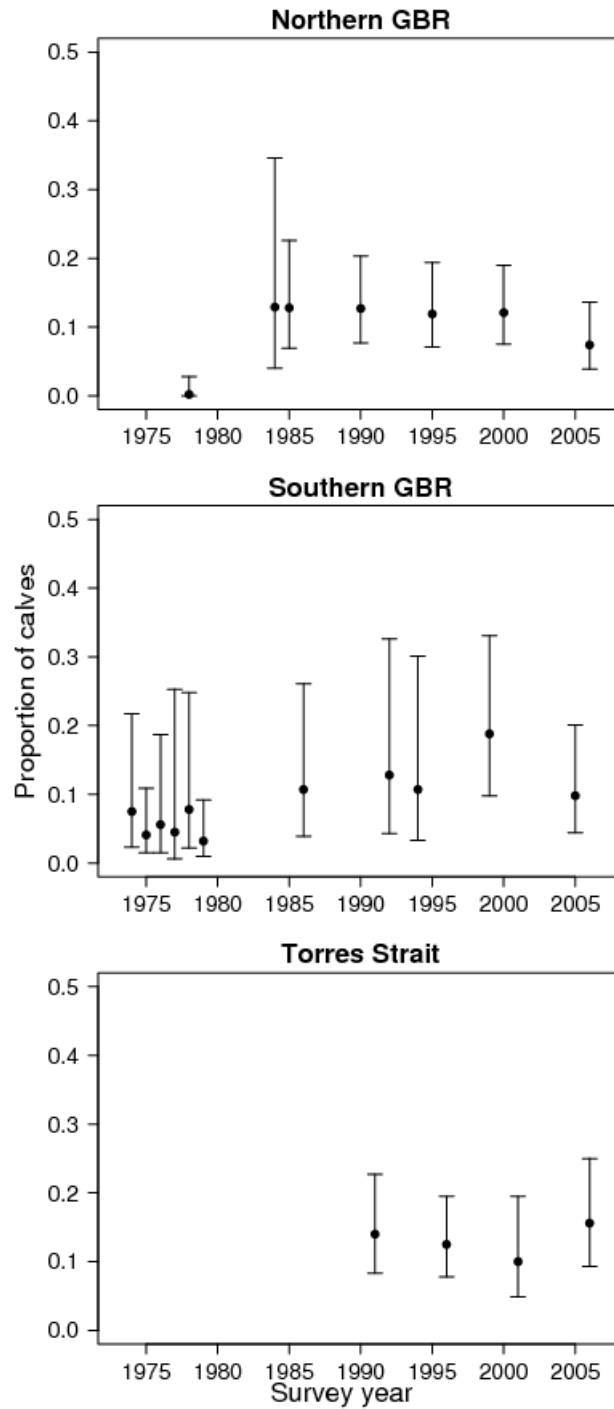


Figure 12 Annual estimates of the proportion of dugong calves for each of the three regions on the east coast of Australia for which the data were available as subregions. Solid points are estimated means; error bars are 95% confidence intervals.

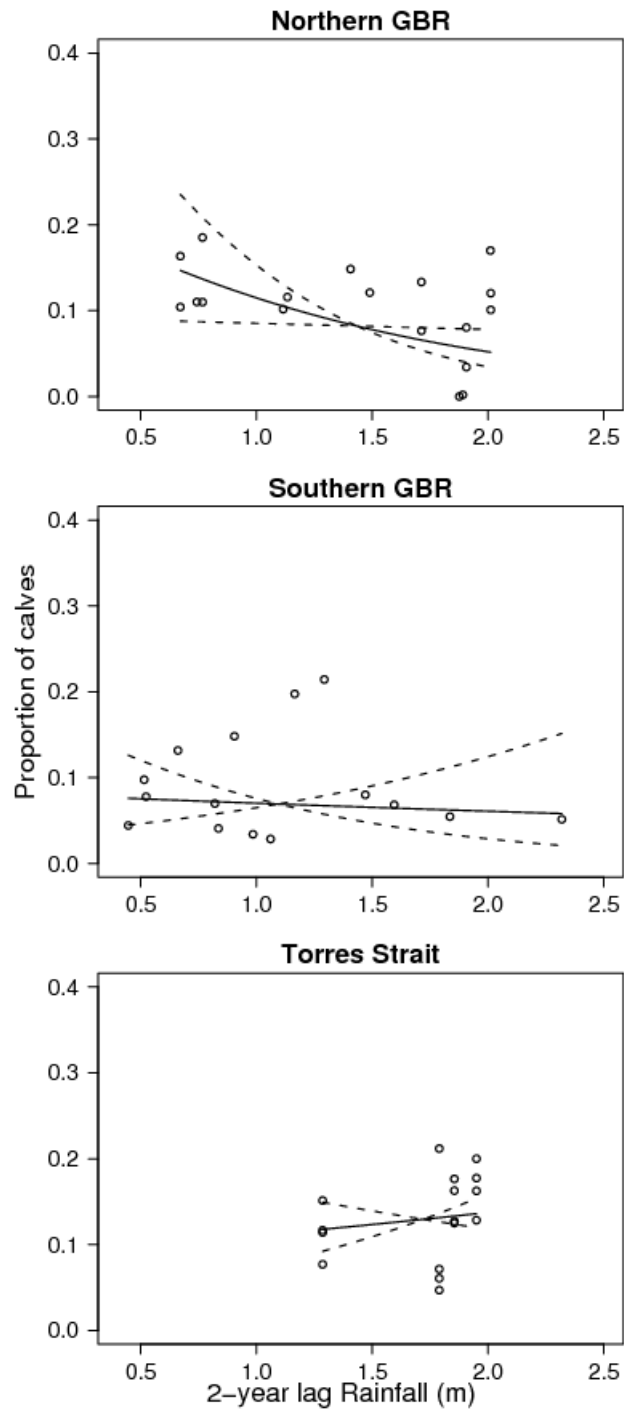


Figure 13. Estimated relationships between the proportion of dugong calves with the total wet season rainfall two years preceding the survey year for the three regions on the east coast of Australia for which data were available in subregions. Solid lines are estimated mean relationship; dashed lines are 95% confidence intervals.

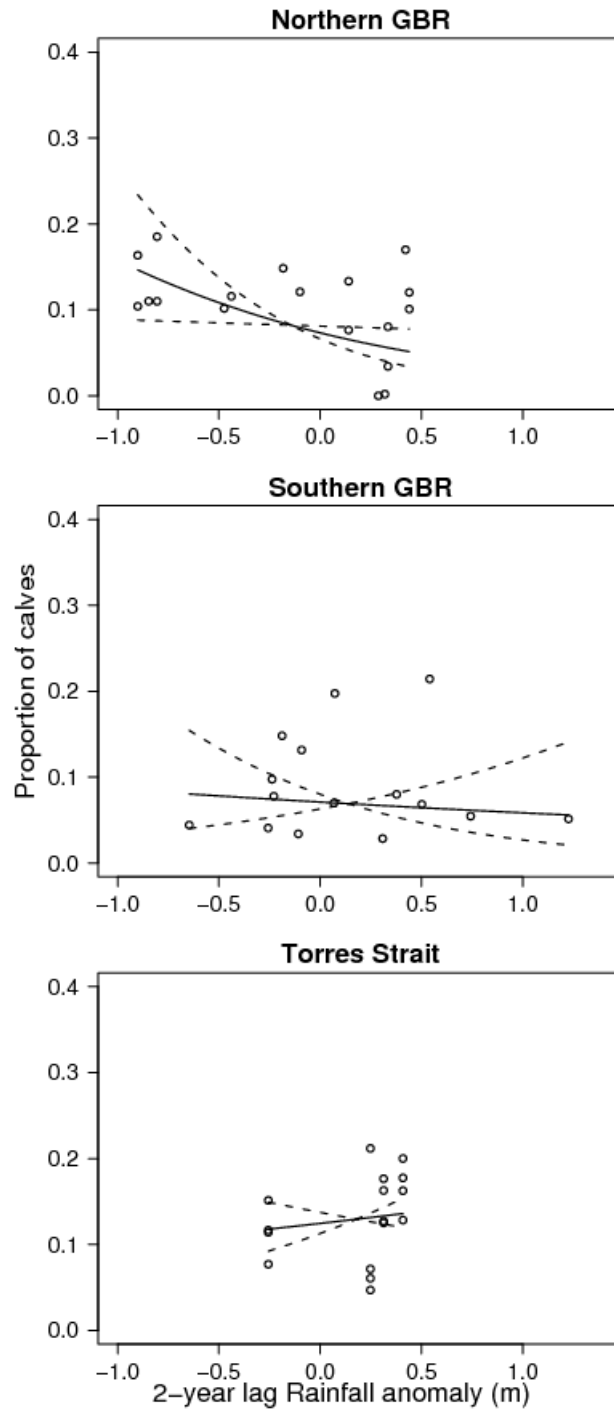


Figure 14. Estimated relationships between the proportion of dugong calves with the rainfall anomalies (m) two years preceding the survey year for the three regions on the east coast of Australia for which data were available in subregions. Solid lines are estimated mean relationship; dashed lines are 95% confidence intervals.

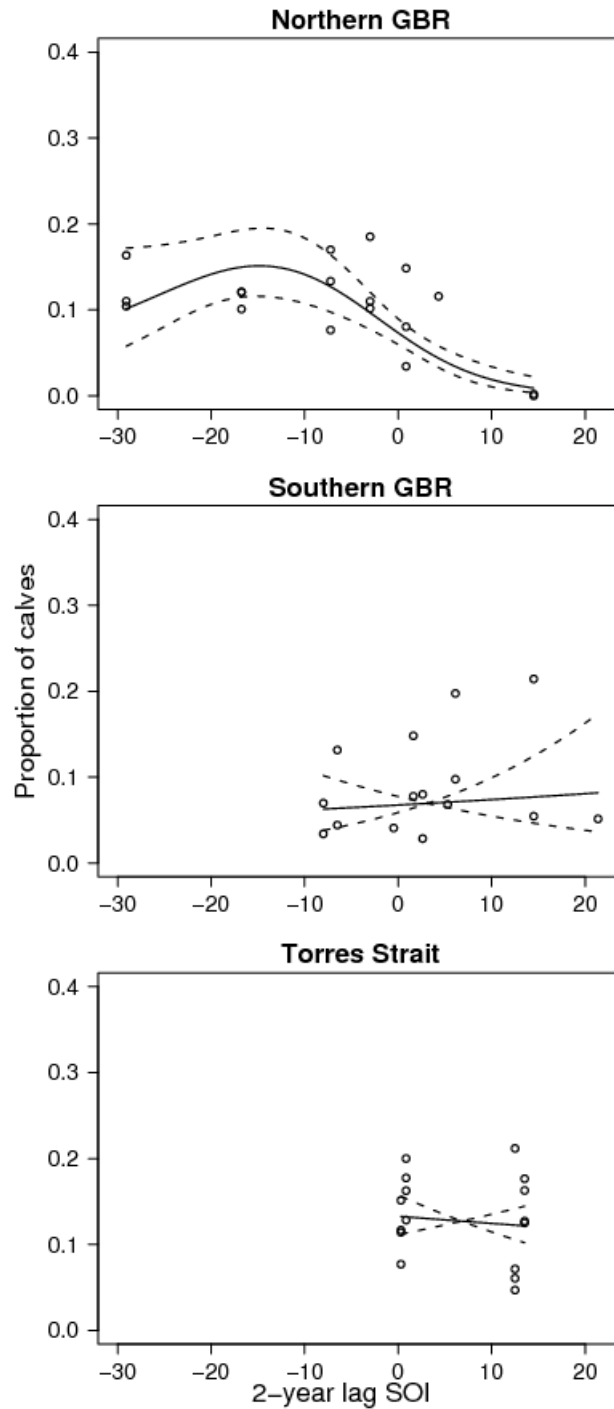


Figure 15 Estimated relationships between the proportion of dugong calves with the Southern Oscillation Index (SOI) two years preceding the survey year for the three regions on the east coast of Australia for which the data were available as subregions. Solid lines are estimated mean relationship; dashed lines are 95% confidence intervals.

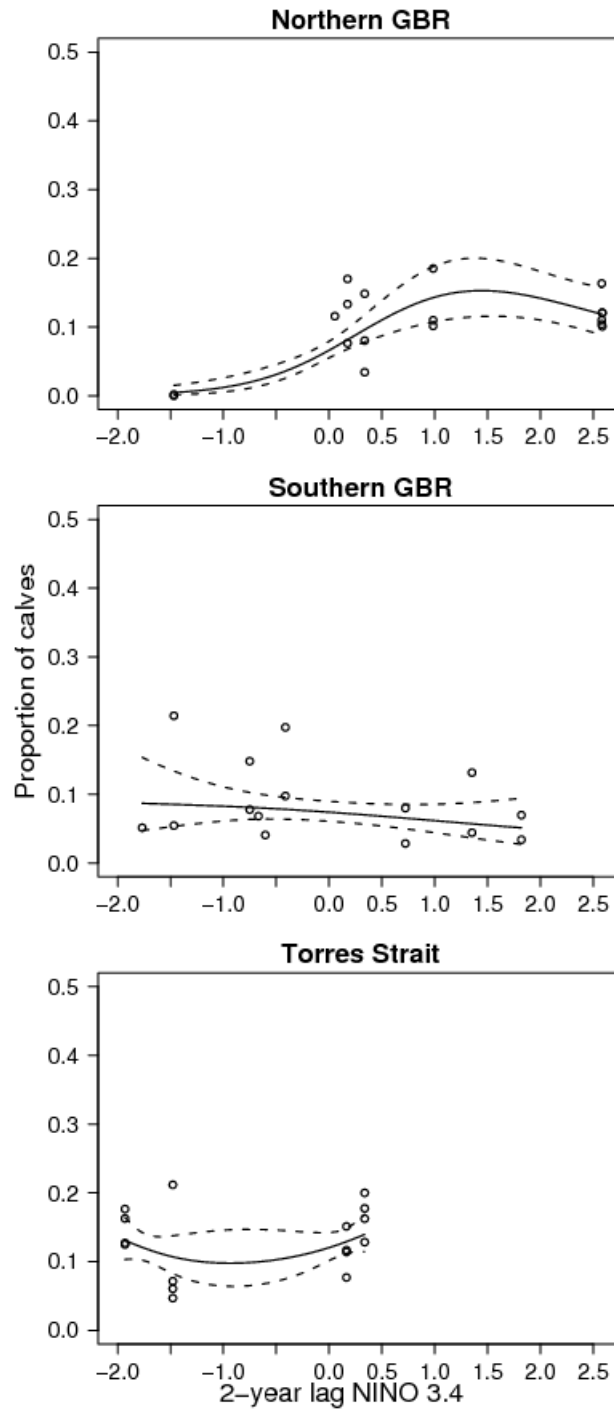
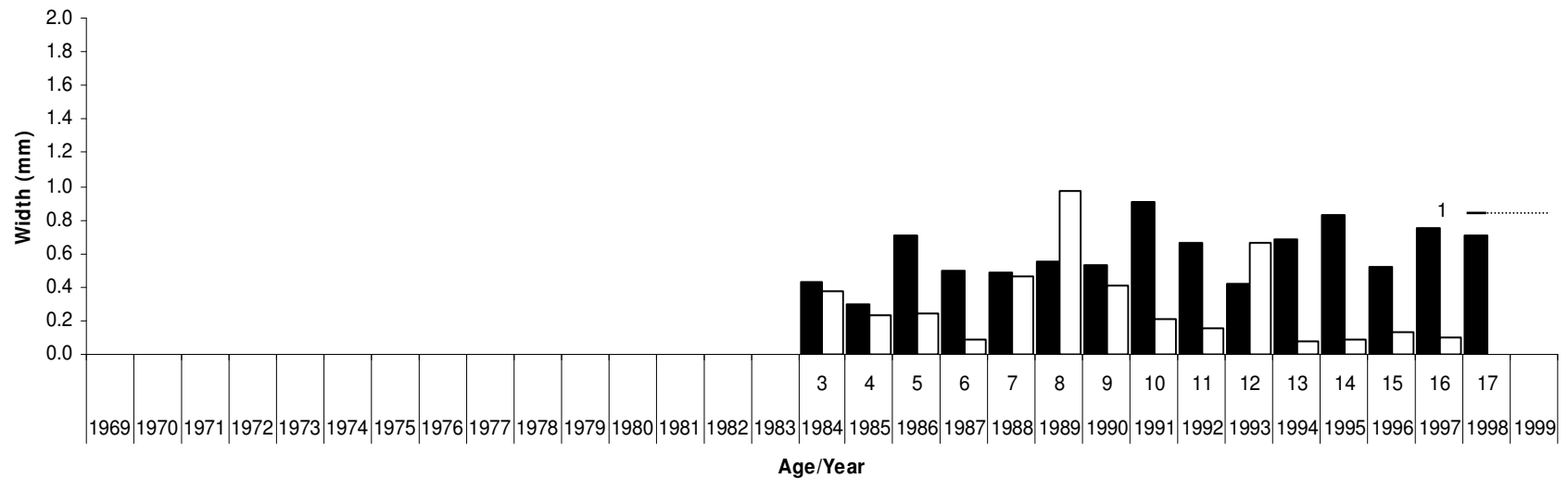


Figure 16 Estimated relationships between the proportion of dugong calves with the NINO 3.4 sea surface temperature index two years preceding the survey year for the three regions on the east coast of Australia for which the data were available as subregions. Solid lines are estimated mean relationship; dashed lines are 95% confidence intervals.

10. Appendix 1

Pattern of spacing of GLGs in dugong tusks. The results are presented graphically for each tusk in the context of known information on dugong life history.

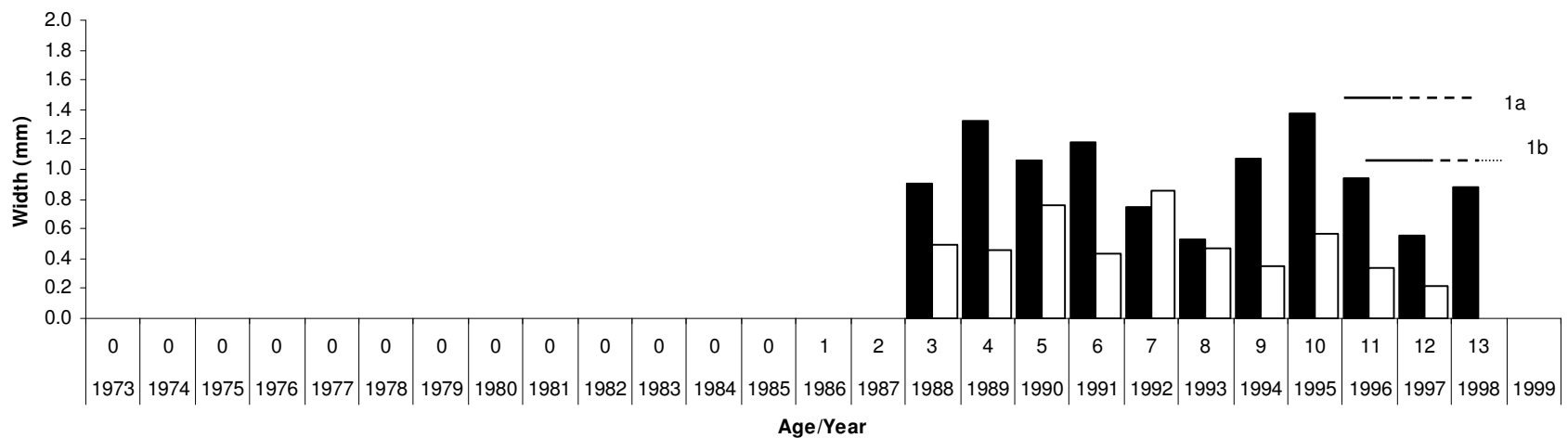


Appendix Figure 1 a. Time series of Growth Layer Groups (Ridge (summer growth/fat deposition): solid bars; Groove (winter growth/fat deposition): empty bars; Pregnant: solid line; Lactating: broken line; dotted line indicates the projected lactation period had the dugong not died). Scenarios are not in any particular order of plausibility, unless stated.

MD19: Female dugong that died in June 1998.

Reproductive history: Pregnant with its first calf when it died.

Scenario 1: The dugong became pregnant with its first calf in the summer of 1998 (indicated by the ridge; age 17) after superior growth/fat deposition the previous year. It died in early pregnancy with a 57.2 cm foetus in June 1998. The dotted line indicates the projected pregnancy had the dugong not died.

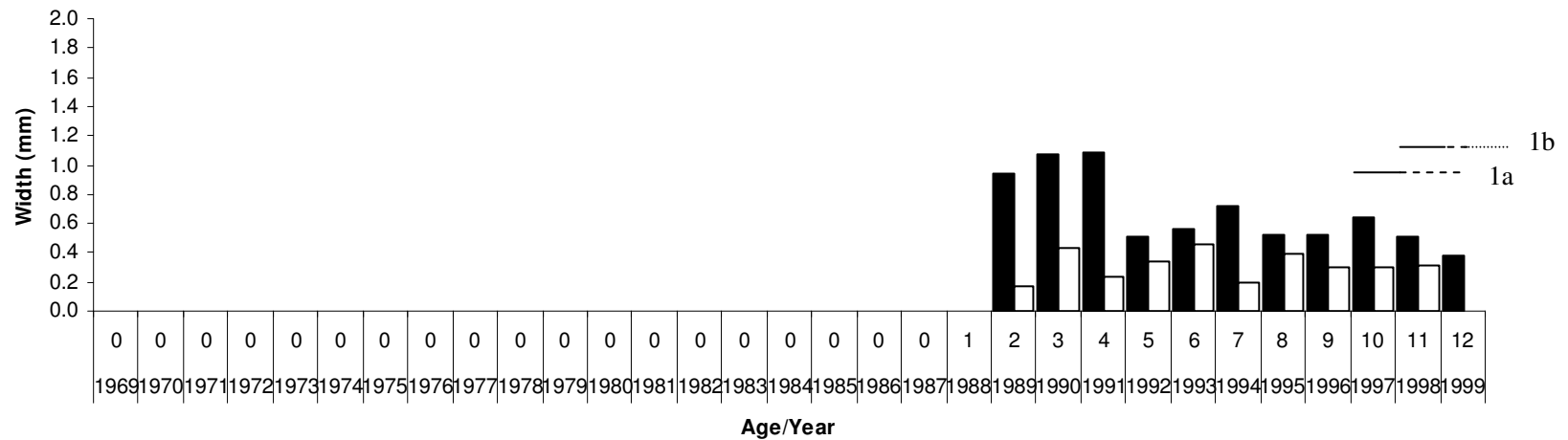


Appendix Figure 1b. Time series of Growth Layer Groups (Ridge (summer growth/fat deposition): solid bars; Groove (winter growth/fat deposition): empty bars; Pregnant: solid line; Lactating: broken line; dotted line indicates the projected lactation period had the dugong not died). Scenarios are not in any particular order of plausibility, unless stated.

MD58: Female dugong that died in August 1998.
Reproductive history: Lactating first calf when died.

Scenario 1a: The dugong became pregnant in the summer of 1996 (indicated by the ridge; age 10) after superior growth/fat deposition the previous year. It gave birth the following summer (age 11) so had reduced growth/fat deposition during the first 12 months of lactation. In the last 6 months of lactation it needed less energy for feeding the calf and therefore grew more/deposited more fat (age 13). We regard this as the more plausible scenario.

Scenario 1b: The dugong became pregnant in the winter (age 11) after superior growth/fat deposition in both the summer and winter of the previous year. It gave birth the following winter (age 12) and did not expend much energy in the second 6 months of lactation when it grew well/fat deposition (age 13); (it died while lactating so the pattern of growth/fat deposition that would have occurred during the last 6 months of lactation is unknown).



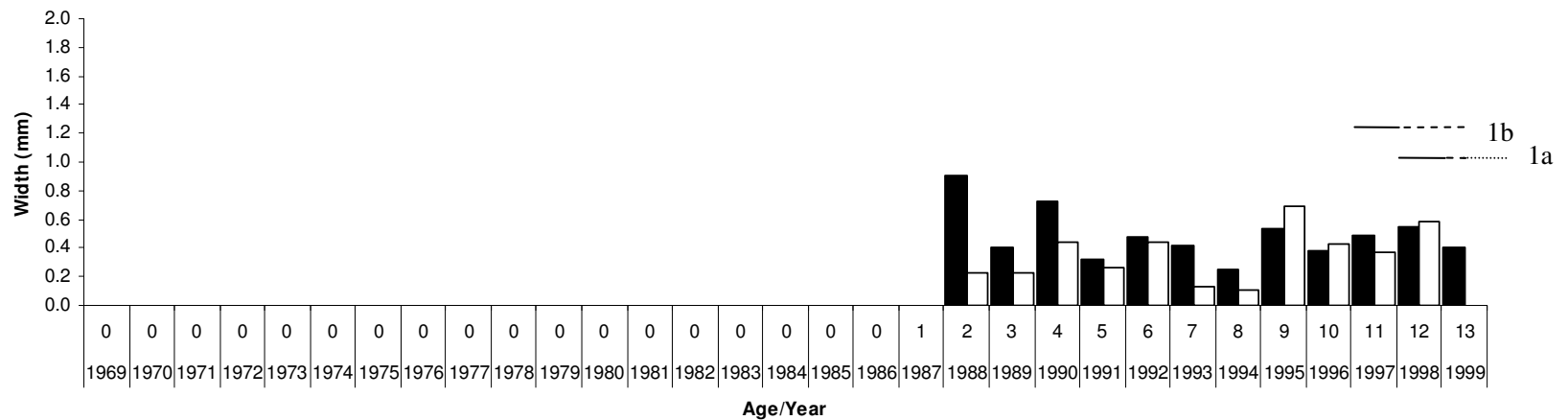
Appendix Figure 1c. Time series of Growth Layer Groups (Ridge (summer growth/fat deposition): solid bars; Groove (winter growth/fat deposition): empty bars; Pregnant: solid line; Lactating: broken line; dotted line indicates the projected lactation period had the dugong not died). Scenarios are not in any particular order of plausibility, unless stated.

MD101: Female dugong that died in March 1999 at age 12.

Reproductive history: Primiparous female lactating her first calf when she died.

Scenario 1a: The dugong grew rapidly until age 5 when growth tapered off. She became pregnant in the summer of 1997 (indicated by the ridge; age 10) during a period of superior growth/fat deposition. She gave birth the following summer and died towards the end of her lactating period. This scenario represents the earliest the dugong could have become pregnant and still be lactating when she died.

Scenario 1b: The dugong became pregnant in the summer of 1998 (indicated by the ridge; age 11) after superior growth/fat deposition the previous year. She gave birth the following summer and died during the early stages of lactating. The dotted line represents the projected lactation period had she not died.



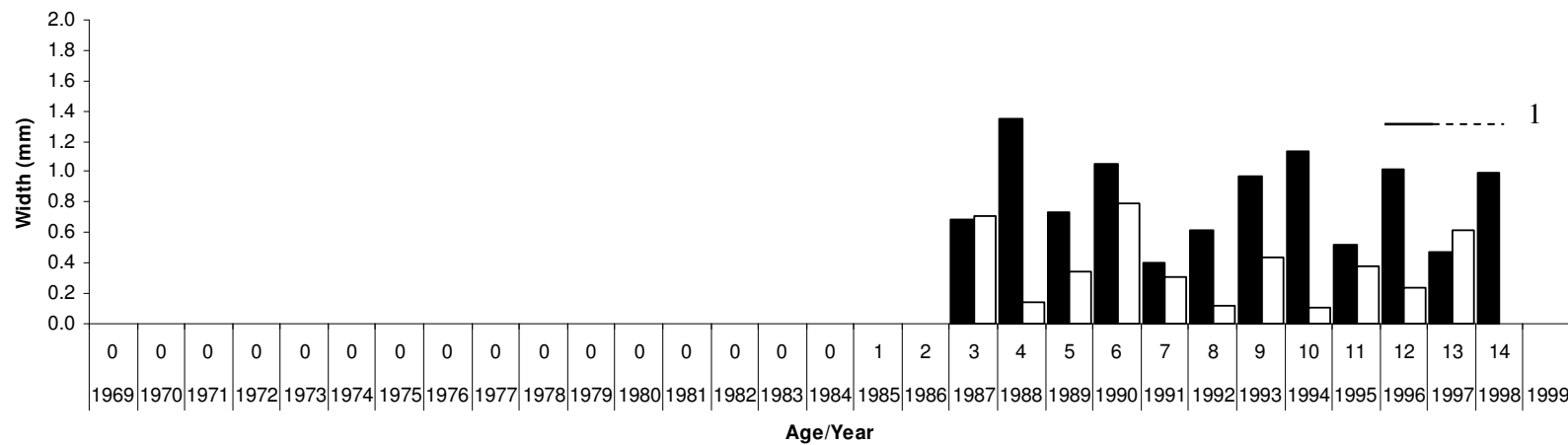
Appendix Figure 1d. Time series of Growth Layer Groups (Ridge (summer growth/fat deposition): solid bars; Groove (winter growth/fat deposition): empty bars; Pregnant: solid line; Lactating: broken line; dotted line indicates the projected lactation period had the dugong not died). Scenarios are not in any particular order of plausibility, unless stated.

MD122: Female dugong that died in April 1999 at age 13.

Reproductive history: Primiparous female lactating her first calf when she died.

Scenario 1a: The dugong became pregnant in the summer of 1998 (indicated by the ridge; age 12) during a period of superior growth/fat deposition. She gave birth the following summer and died during the early stages of lactating. The dotted line represents the projected lactating period had she not died.

Scenario 1b: The dugong became pregnant in the summer of 1997 (indicated by the ridge; age 11). She gave birth the following summer (age 12) and died at the end of her lactating period. We regard this scenario as less plausible than scenario 1a because we would expect growth to be low during the winter of 1998 (indicated by the groove; age 12) because of the energetic demands of the start of lactation. Instead, there is superior growth/fat deposition at this time. The good growth/fat deposition in the winter of 1998 could indicate very good environmental conditions throughout the whole year.

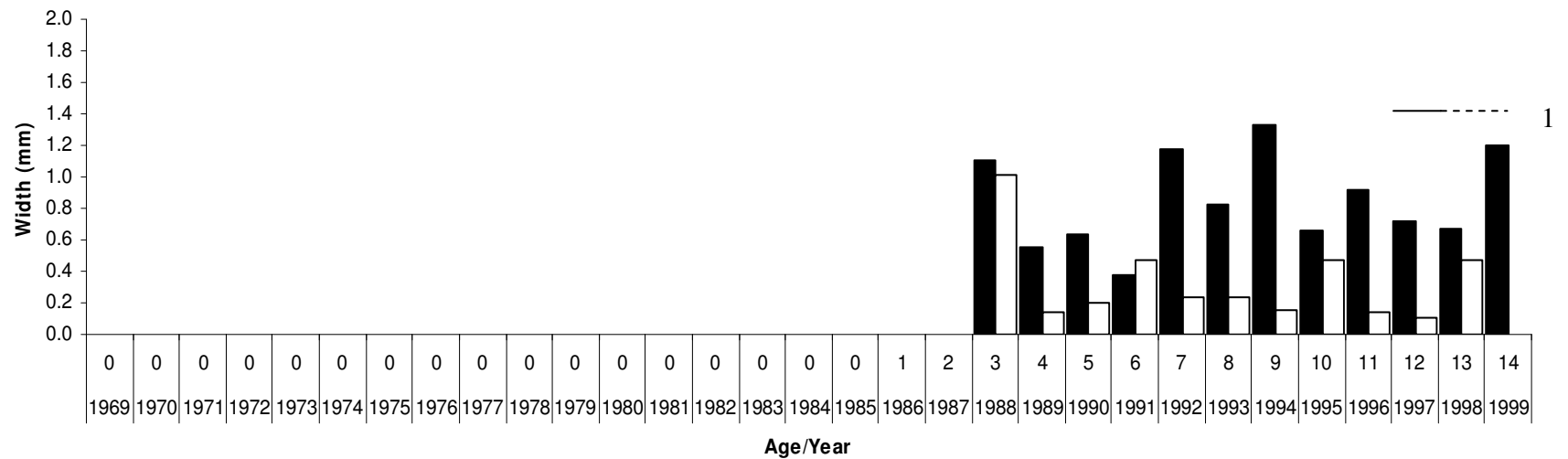


Appendix Figure 1e. Time series of Growth Layer Groups (Ridge (summer growth/fat deposition): solid bars; Groove (winter growth/fat deposition): empty bars; Pregnant: solid line; Lactating: broken line; dotted line indicates the projected lactation period had the dugong not died). Scenarios are not in any particular order of plausibility, unless stated.

MD73: Female dugong that died in September 1998 at age 14.

Reproductive history: Primiparous female lactating her first calf when she died.

Scenario 1a: The dugong became pregnant in the summer of 1996 (indicated by the ridge; age 12) during a period of superior growth/fat deposition. She gave birth the following summer and died during the late stages of lactating.

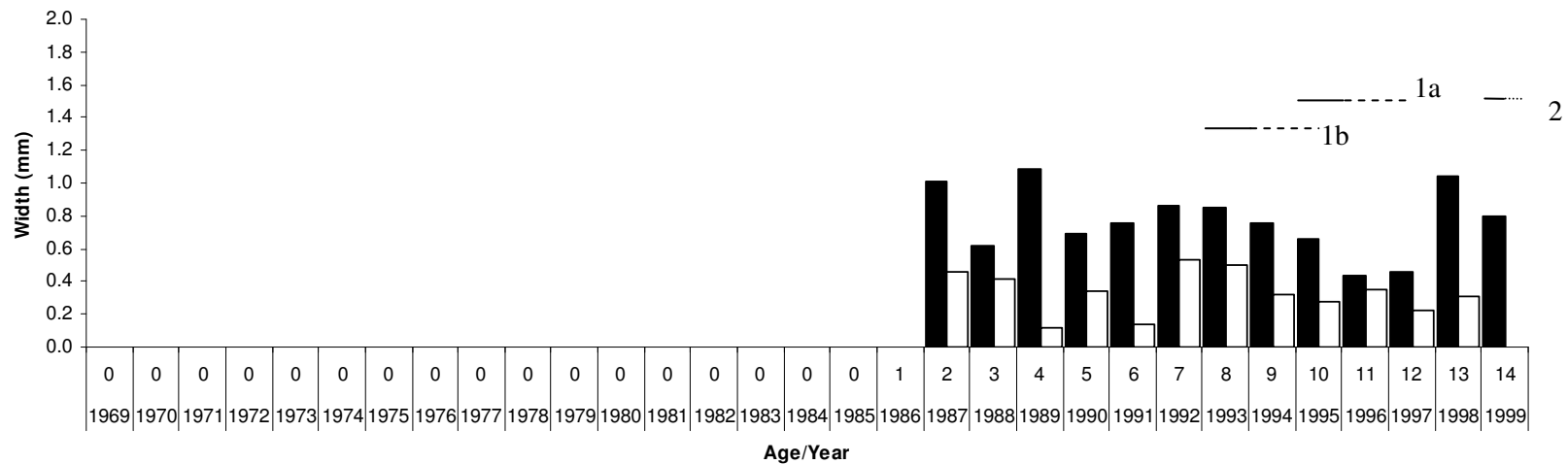


Appendix Figure 1f. Time series of Growth Layer Groups (Ridge (summer growth/fat deposition): solid bars; Groove (winter growth/fat deposition): empty bars; Pregnant: solid line; Lactating: broken line; dotted line indicates the projected lactation period had the dugong not died). Scenarios are not in any particular order of plausibility, unless stated.

MD155: Female dugong that died in July 1999 at age 14.

Reproductive history: Primiparous female lactating her first calf when she died.

Scenario 1a: The dugong became pregnant in the summer of 1997 (indicated by the ridge; age 12) during a period of superior growth/fat deposition. She gave birth the following summer and experienced lower growth/fat deposition during the early stages of lactation, but grew better/had better fat deposition in the last part of lactation, during which she died.



Appendix Figure 1g. Time series of Growth Layer Groups (Ridge (summer growth/fat deposition): solid bars; Groove (winter growth/fat deposition): empty bars; Pregnant: solid line; Lactating: broken line; dotted line indicates the projected lactation period had the dugong not died). Scenarios are not in any particular order of plausibility, unless stated.

MD108: Female dugong that died in April 1999 at age 14.

Reproductive history: Multiparous female in the early stages of pregnancy (foetus 30.08cm) with her second calf.

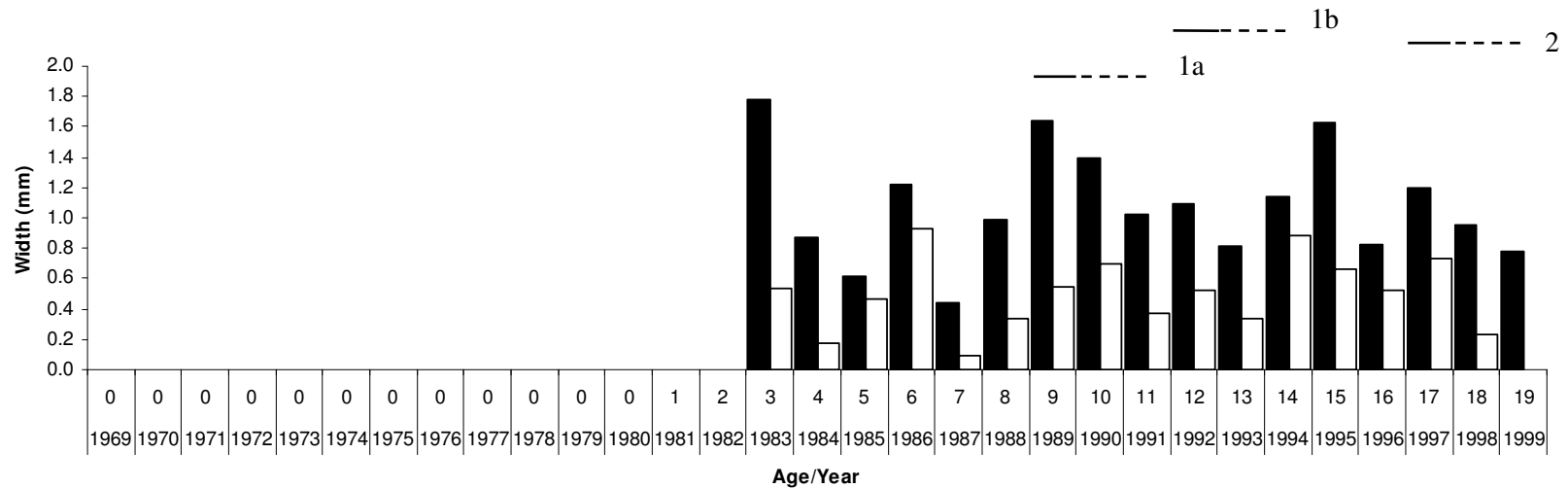
Second calf:

Scenario 2: The dugong became pregnant in the summer of 1999 (indicated by the ridge; age 14) after good growth/fat deposition the previous year. She died during the early stages of pregnancy and the dotted line indicates the projected pregnancy period .

First calf:

Scenario 1a: The dugong became pregnant in the summer of 1995(indicated by the ridge, age 10) after superior growth/fat deposition in the preceding few years. She gave birth in the summer of 1996 (age 11) and grew more slowly/had less fat deposition during 18 months of lactation, after which she started to deposit fat again. We regard this as the more likely scenario because it assumes that there was lower growth/fat deposition during lactation than scenario1b.

Scenario 1b: The dugong became pregnant in the summer of 1993 (indicated by the ridge; age 8) after superior growth/fat deposition the previous year. She gave birth in the summer of 1994 (age 9) and grew less/had less fat deposition during lactation.



Appendix Figure 1h. Time series of Growth Layer Groups (Ridge (summer growth/fat deposition): solid bars; Groove (winter growth/fat deposition): empty bars; Pregnant: solid line; Lactating: broken line; dotted line indicates the projected lactation period had the dugong not died). Scenarios are not in any particular order of plausibility, unless stated.

MD138: Female dugong that died in May 1999 at age 19.

Reproductive history: Multiparous female lactating her second calf.

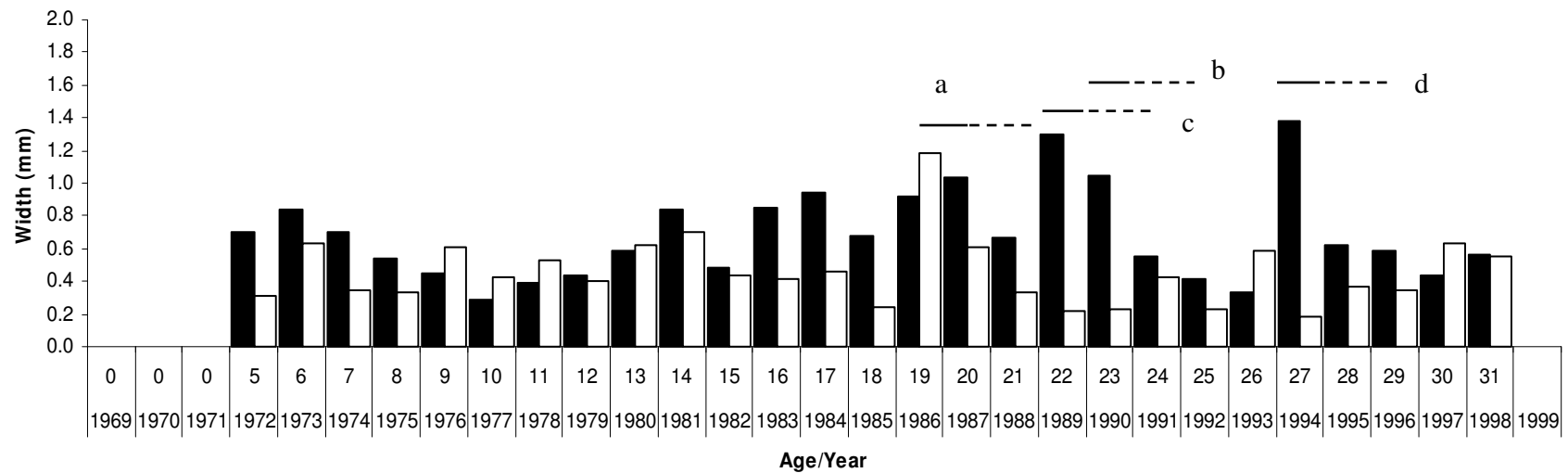
Second calf:

Scenario 2. The dugong became pregnant in the summer of 1997 (indicated by the ridge; age 17). She gave birth the following summer (age 18) and experienced slow growth/fat deposition in the first 12 months of lactation.

First calf:

Scenario 1a. The dugong became pregnant in the summer of 1989 (indicated by the ridge; age 9). She gave birth the following summer and experienced less growth/fat deposition during lactation.

Scenario 1b. The dugong became pregnant in the summer of 1992 (indicated by the ridge; age 12). She gave birth the following summer and experienced less growth/fat deposition during lactation.



Appendix Figure 1i. Time series of Growth Layer Groups (Ridge (summer growth/fat deposition): solid bars; Groove (winter growth/fat deposition): empty bars; Pregnant: solid line; Lactating: broken line; dotted line indicates the projected lactation period had the dugong not died). Scenarios are not in any particular order of plausibility, unless stated.

MD56: Female dugong that died in August 1998 at age 31.

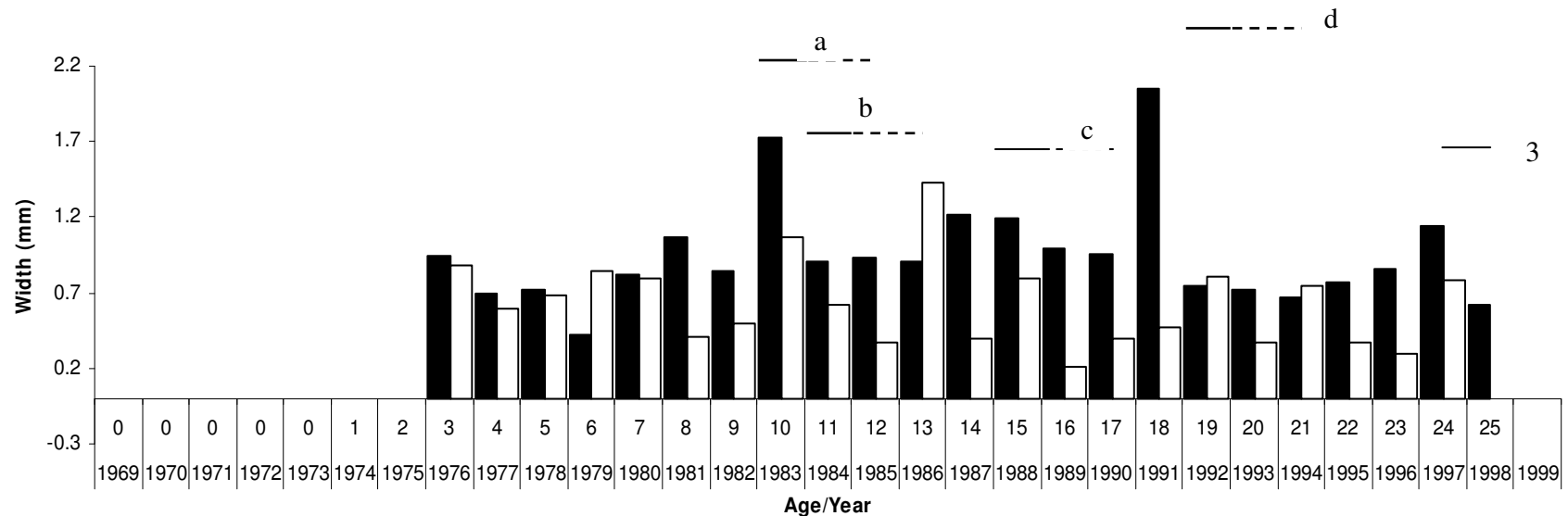
Reproductive history: Multiparous female that is still reproductively active and has had 2 calves.

Scenario a. The dugong became pregnant in the winter of 1986 (indicated by the groove; age 19) during superior growth that year. It gave birth the following winter (age 20) and experienced lower growth/fat deposition lactation.

Scenario b. The dugong became pregnant in the summer of 1990 (indicated by the ridge; age 23) after superior growth/fat deposition the previous summer. She gave birth the following summer (age 24) and experienced low growth/fat deposition during lactation.

Scenario c. The dugong became pregnant in the summer of 1989 (indicated by the ridge; age 22) during a period of superior growth/fat deposition. She gave birth the following summer (age 23) and experienced low growth/fat deposition during lactation. We regard this as a less plausible scenario than (b) because growth/fat deposition did not drop off as sharply for the start of lactation.

Scenario d. The dugong became pregnant in the summer of 1994 (indicated by the ridge; age 27) during a period of superior growth/fat deposition. She gave birth the following summer (age 28) and experienced low growth/fat deposition during lactation.



Appendix Figure 1j. Time series of Growth Layer Groups (Ridge (summer growth/fat deposition): solid bars; Groove (winter growth/fat deposition): empty bars; Pregnant: solid line; Lactating: broken line; dotted line indicates the projected lactation period had the dugong not died). Scenarios are not in any particular order of plausibility, unless stated.

MD60: Female dugong that died in August 1998 at age 25.

Reproductive history: Multiparous female in the late stages of pregnancy (foetus 105.2cm) with her third calf.

Third calf:

Scenario 3: The dugong became pregnant in the summer/autumn of 1997 (indicated by the ridge; age 24) during a period of superior growth/fat deposition. She was in the very late stages of pregnancy in the winter 1998 (age 25) when she died.

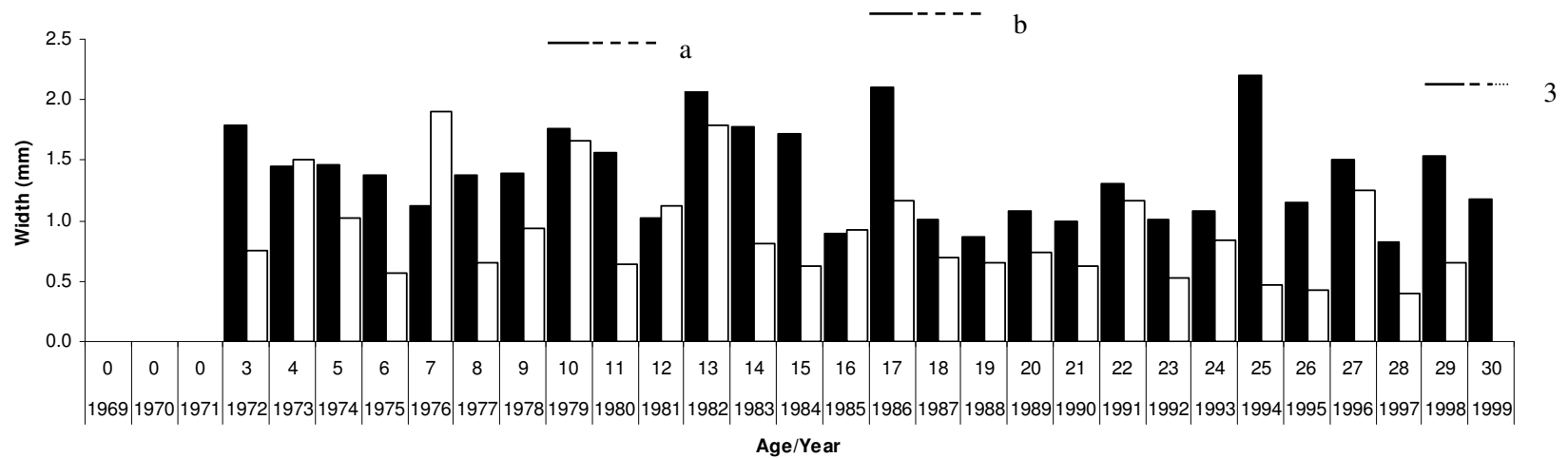
Other 2 calves:

Scenario a. The dugong became pregnant in the summer of 1983 (indicated by the ridge; age 10) during a period of superior growth/fat deposition. She gave birth the following summer (age 11) and experienced less growth/fat deposition during lactation.

Scenario b. The dugong became pregnant in the summer of 1984 (indicated by the ridge; age 11) after a period of superior growth/fat deposition the previous year. She gave birth the following summer (age 12) and experienced low growth/fat deposition during lactation, particularly in the winter.

Scenario c: The dugong became pregnant in the summer of 1988 (indicated by the ridge; age 15) during a period of reasonably good growth/fat deposition. She gave birth the following summer (age 16) and experienced less growth/fat deposition during lactation, particularly in the winter.

Scenario d: The dugong became pregnant in the summer of 1992 (indicated by the ridge; age 15) after a period of superior growth/fat deposition the previous year. She gave birth the following summer (age 20) and experienced less growth/fat deposition in the first 6 months of lactation, after which she had better growth/fat deposition.



Appendix Figure 1k. Time series of Growth Layer Groups (Ridge (summer growth/fat deposition): solid bars; Groove (winter growth/fat deposition): empty bars; Pregnant: solid line; Lactating: broken line; dotted line indicates the projected lactation period had the dugong not died). Scenarios are not in any particular order of plausibility, unless stated.

MD161: Female dugong that died in May 1999 at age 30.

Reproductive history: Multiparous female that is lactating her third calf.

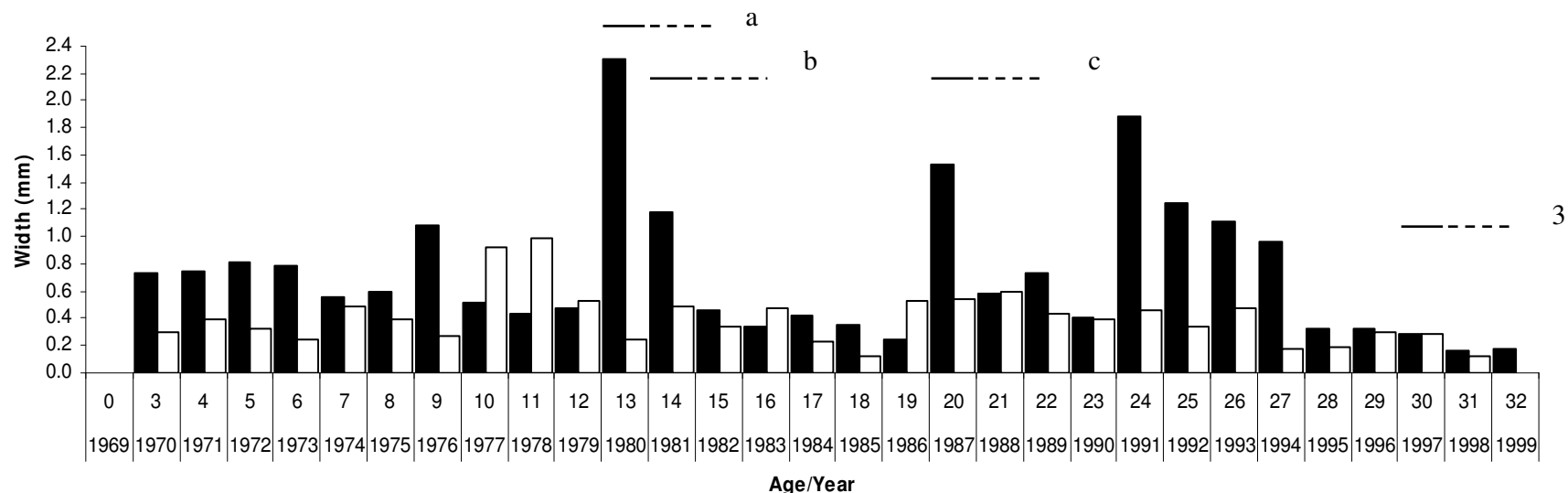
Third calf:

Scenario 3: The dugong became pregnant in the summer of 1998 (indicated by the ridge; age 29) during a period of superior growth/fat deposition. It gave birth the following summer and experienced lower growth during lactation. It died during the first 6 months of lactation and the dotted line indicates the projected lactation period if the dugong had not died.

Other calves:

Scenario a. The dugong became pregnant in the summer of 1979 (indicated by the ridge; age 10) during a period of superior growth/fat deposition. She gave birth the following summer (age 11) and experienced lower growth/fat deposition during lactation.

Scenario b. The dugong became pregnant in the summer of 1986 (indicated by the ridge; age 17) during a period of superior growth/fat deposition. She gave birth the following summer (age 18) and experienced lower growth/fat deposition during lactation.



Appendix Figure 11. Time series of Growth Layer Groups (Ridge (summer growth/fat deposition): solid bars; Groove (winter growth/fat deposition): empty bars; Pregnant: solid line; Lactating: broken line; dotted line indicates the projected lactation period had the dugong not died). Scenarios are not in any particular order of plausibility, unless stated.

MD148: Female dugong that died in July 1999 at age 32.

Reproductive history: Multiparous female that is lactating its third calf.

Third calf:

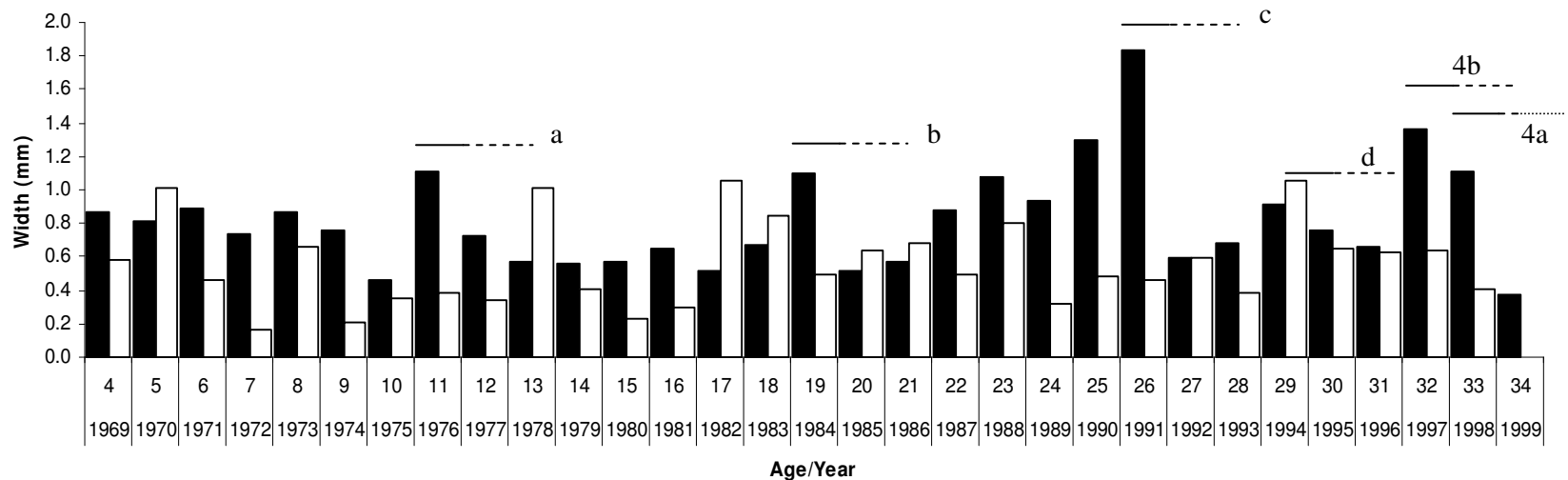
Scenario 3: The dugong became pregnant in the summer of 1997 (indicated by the ridge; age 30). It gave birth the following summer and experienced lower growth during lactation. It died at the end of its lactating period.

Other calves:

Scenario a. The dugong became pregnant in the summer of 1980 (indicated by the ridge; age 13) during good growth/fat deposition. She gave birth the following summer (age 14) and experienced lower growth/fat deposition during lactation.

Scenario b. The dugong became pregnant in the summer of 1981 (indicated by the ridge; age 11) during a period of superior growth/fat deposition in that year and the previous year. She gave birth the following summer (age 12) and experienced lower growth/fat deposition during lactation.

Scenario c. The dugong became pregnant in the summer of 1987 (indicated by the ridge; age 20) during a period of superior growth/fat deposition. She gave birth the following summer (age 21) and experienced lower growth/fat deposition during lactation.



Appendix Figure 1m. Time series of Growth Layer Groups (Ridge (summer growth/fat deposition): solid bars; Groove (winter growth/fat deposition): empty bars; Pregnant: solid line; Lactating: broken line; dotted line indicates the projected lactation period had the dugong not died). Scenarios are not in any particular order of plausibility, unless stated.

MD98: Female dugong that died in March 1999 at age 34.

Reproductive history: Multiparous female was lactating its fourth calf when it died. The fourth calf was with the mother when it died.

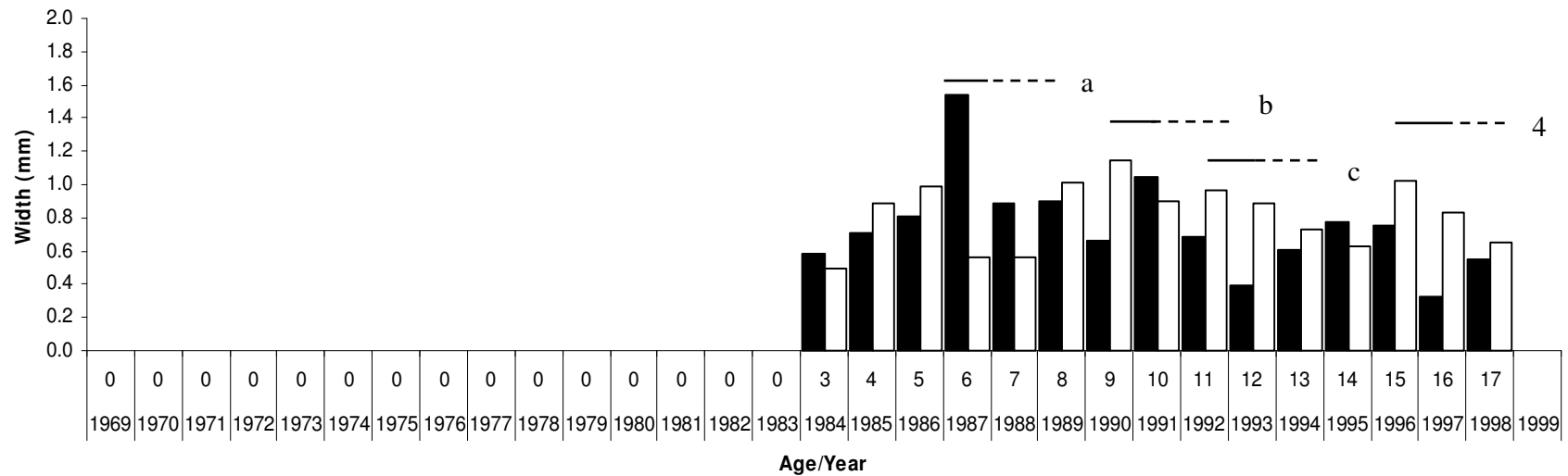
Fourth calf:

Scenario 4a. The dugong became pregnant with its fourth calf in the summer of 1998 (indicated by the ridge; age 33) after a period of superior growth/fat deposition the previous year. It gave birth the following summer (age 34) and was lactating the calf when it died so growth/fat deposition for the last 12 months of lactation is unknown. We consider this the most plausible scenario.

Scenario 4b. The dugong became pregnant with its fourth calf in the summer of 1997 (indicated by the ridge; age 32) while experiencing a period of superior growth/fat deposition. It gave birth the following summer and (age 33) and growth/fat deposition slowed during lactation. It was at the end of the lactation period when it died.

Other 3 calves: There are several possible scenarios and we consider these four the most plausible.

- a. The dugong became pregnant in the summer of 1976 (indicated by the ridge; age 11) during a period of superior growth/fat deposition. It gave birth the following summer (age 12) and experienced poor growth/fat deposition during lactation.
- b. The dugong became pregnant in the summer of 1984 (indicated by the ridge; age 19) during a period of superior growth/fat deposition and after good growth/fat deposition the previous year. It gave birth the following summer (age 20) and experienced poorer, but increasing growth/fat deposition during lactation. After finishing suckling its calf its growth/fat deposition improved again (indicated by the ridge; age 22).
- c. The dugong became pregnant in the summer of 1991 (indicated by the ridge; age 26) during a period of superior growth/fat deposition and after several years of increasingly superior growth/fat deposition. It gave birth the following summer and grew poorly/had poor fat deposition during lactation. After finishing suckling its calf it grew well/had superior fat deposition again (indicated by the ridge; age 29).
- d. The dugong became pregnant in the winter of 1994 (indicated by the groove; age 29) during a period of superior growth/fat deposition and after superior growth/fat deposition the previous summer. It gave birth the following winter and grew poorly/had poor fat deposition during lactation. After finishing suckling its calf it grew well/had superior fat deposition again (indicated by the ridge; age 32).



Appendix Figure 1n. Time series of Growth Layer Groups (Ridge (summer growth/fat deposition): solid bars; Groove (winter growth/fat deposition): empty bars; Pregnant: solid line; Lactating: broken line; dotted line indicates the projected lactation period had the dugong not died). Scenarios are not in any particular order of plausibility, unless stated.

MD96: Female dugong that died in November 1998 at age 17.

Reproductive history: Multiparous female that was lactating its fourth calf.

Fourth calf:

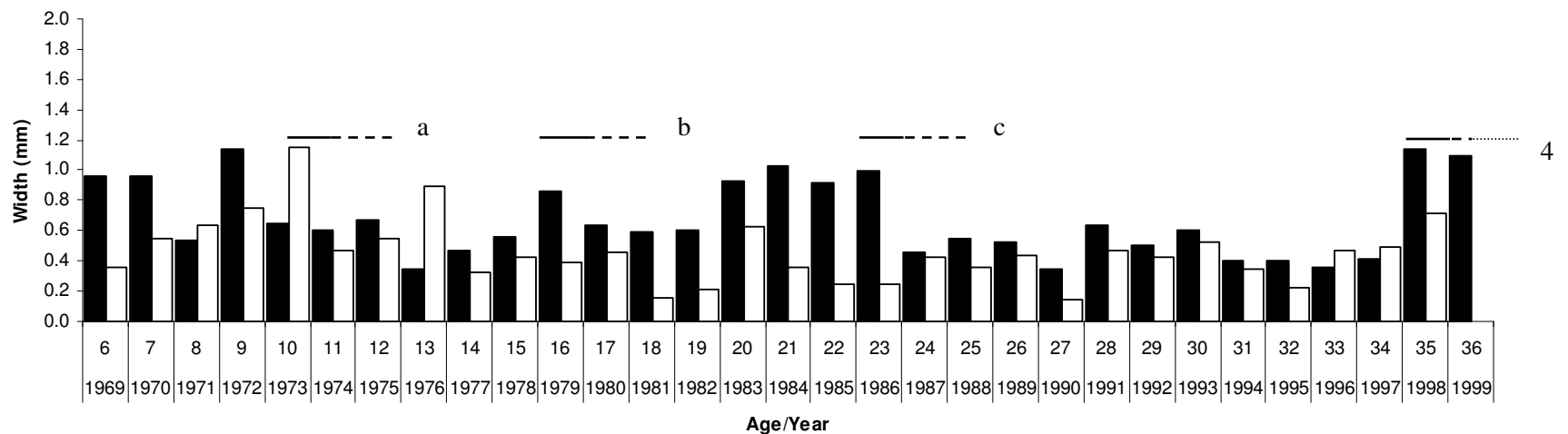
Scenario 4: The dugong became pregnant in the winter of 1996 (indicated by the groove; age 15). It gave birth the following winter and experienced lower growth/fat deposition during lactation. It died at the end of its lactating period.

Other calves:

Scenario a. The dugong became pregnant in the summer of 1987 (indicated by the ridge; age 6) during a period of superior growth/fat deposition. She gave birth the following summer (age 7) and experienced lower growth/fat deposition during lactation.

Scenario b. The dugong became pregnant in the winter of 1990 (indicated by the groove; age 9) during good growth/fat deposition. She gave birth the following winter (age 10) and experienced lower growth/fat deposition during lactation, which improved towards the end of the lactating period when she became pregnant again.

Scenario c. The dugong became pregnant in the winter of 1992 (indicated by the groove; age 11) during good growth/fat deposition and lactating a previous calf. She gave birth the following winter (age 12) and experienced lower growth/fat deposition during lactation.



Appendix Figure 10. Time series of Growth Layer Groups (Ridge (summer growth/fat deposition): solid bars; Groove (winter growth/fat deposition): empty bars; Pregnant: solid line; Lactating: broken line; dotted line indicates the projected lactation period had the dugong not died). Scenarios are not in any particular order of plausibility, unless stated.

Figure x. **MD124:** Female dugong that died in April 1999 at age 36.

Reproductive history: Multiparous female that was lactating its fourth calf.

Fourth calf:

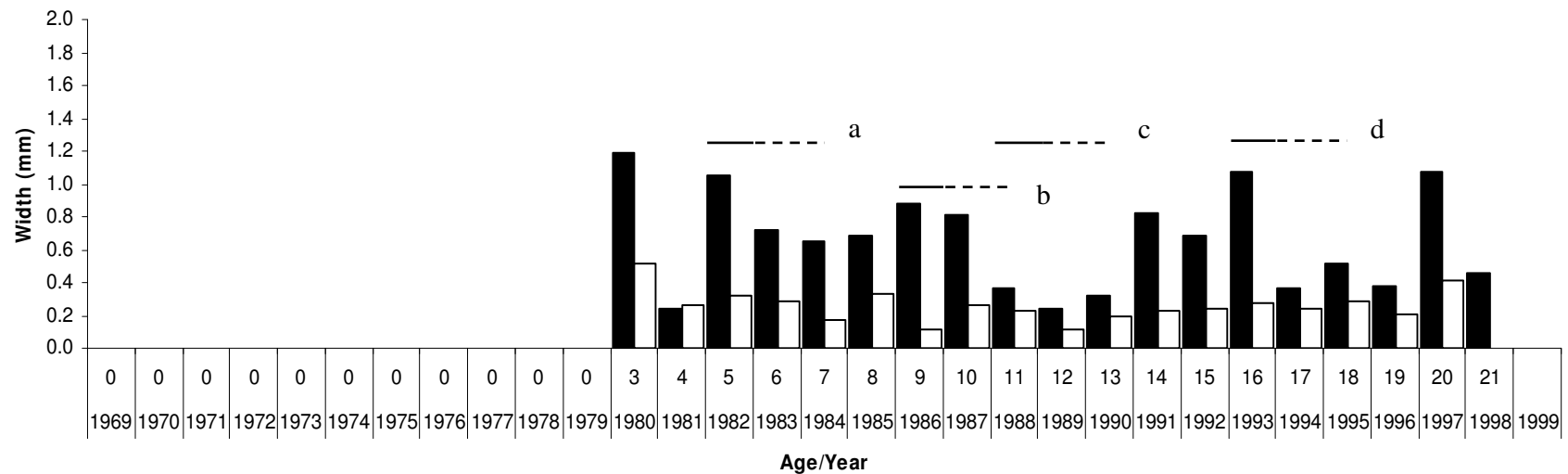
Scenario 4: The dugong became pregnant in the summer of 1998 (indicated by the ridge; age 35). It gave birth the following winter and experienced lower growth during lactation. It had slightly lower growth/fat deposition in the first part of lactation, but died early on in its lactating period so growth/fat deposition during lactation is unknown.

Other calves:

Scenario a. The dugong became pregnant in the winter of 1973 (indicated by the groove; age 10) during a period of superior growth/fat deposition. She gave birth the following winter (age 11) and experienced lower growth/fat deposition during lactation.

Scenario b. The dugong became pregnant in the summer of 1979 (indicated by the ridge; age 16) during a period of superior growth/fat deposition. She gave birth the following summer (age 17) and experienced lower growth/fat deposition during lactation.

Scenario c. The dugong became pregnant in the summer of 1986 (indicated by the ridge; age 23) during a period of superior growth/fat deposition. She gave birth the following winter (age 24) and experienced lower growth/fat deposition during lactation.



Appendix Figure 1p. Time series of Growth Layer Groups (Ridge (summer growth/fat deposition): solid bars; Groove (winter growth/fat deposition): empty bars; Pregnant: solid line; Lactating: broken line; dotted line indicates the projected lactation period had the dugong not died). Scenarios are not in any particular order of plausibility, unless stated.

MD41: Female dugong that died in July 1998 at age 21.

Reproductive history: Multiparous female that is still reproductively active and has had 4 calves.

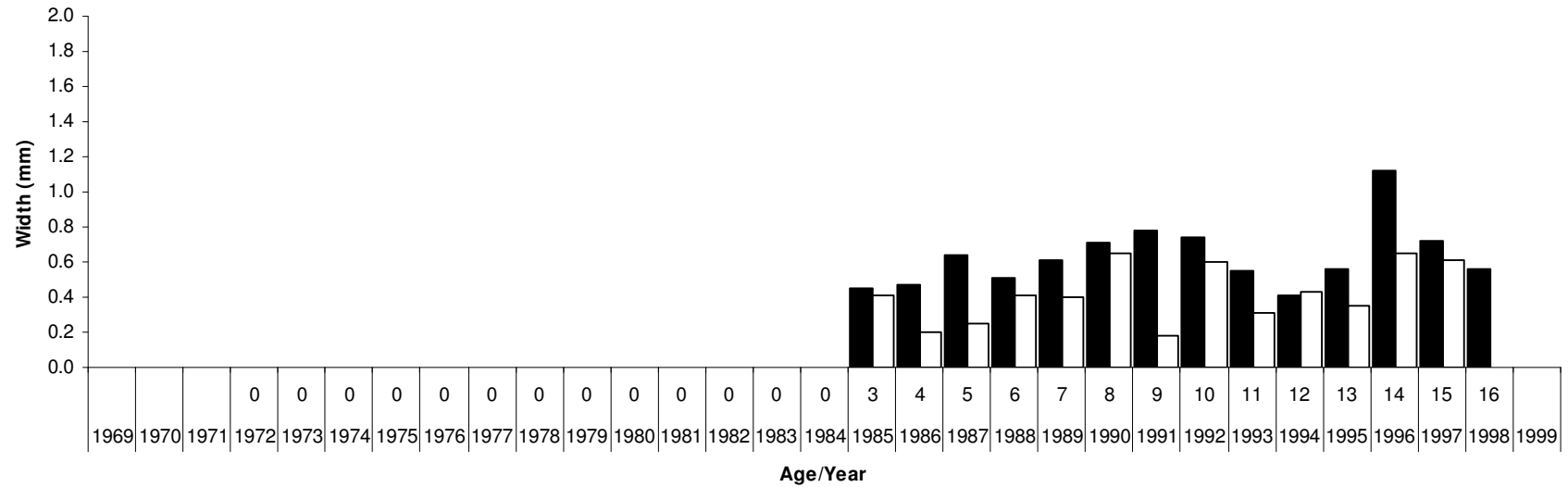
Scenario a. The dugong became pregnant in the summer of 1982 (indicated by the ridge (age 5) during a period of superior growth/fat deposition. She gave birth the following summer (age 6) and experienced lower growth/fat deposition. She was still growing/depositing fat reasonably well, probably because she was young.

Scenario b. The dugong became pregnant in the summer of 1986 (age 9) after a period of superior growth/fat deposition the previous few years. She gave birth the following summer (10) and was still in good condition during the first few months of lactation, after which she had lower growth/fat deposition.

Scenario c. The dugong became pregnant in the summer of 1988 (age 11) after a period of superior growth/fat deposition the previous few years and while still suckling her previous calf. She gave birth the following summer (age 12) and experienced low growth/fat deposition during lactation.

Scenario d. The dugong became pregnant in the summer of 1993 (indicated by the ridge; age 16) during a period of superior growth that year and the previous 2 years. She gave birth the following summer (age 17) and experienced low growth/fat deposition during lactation.

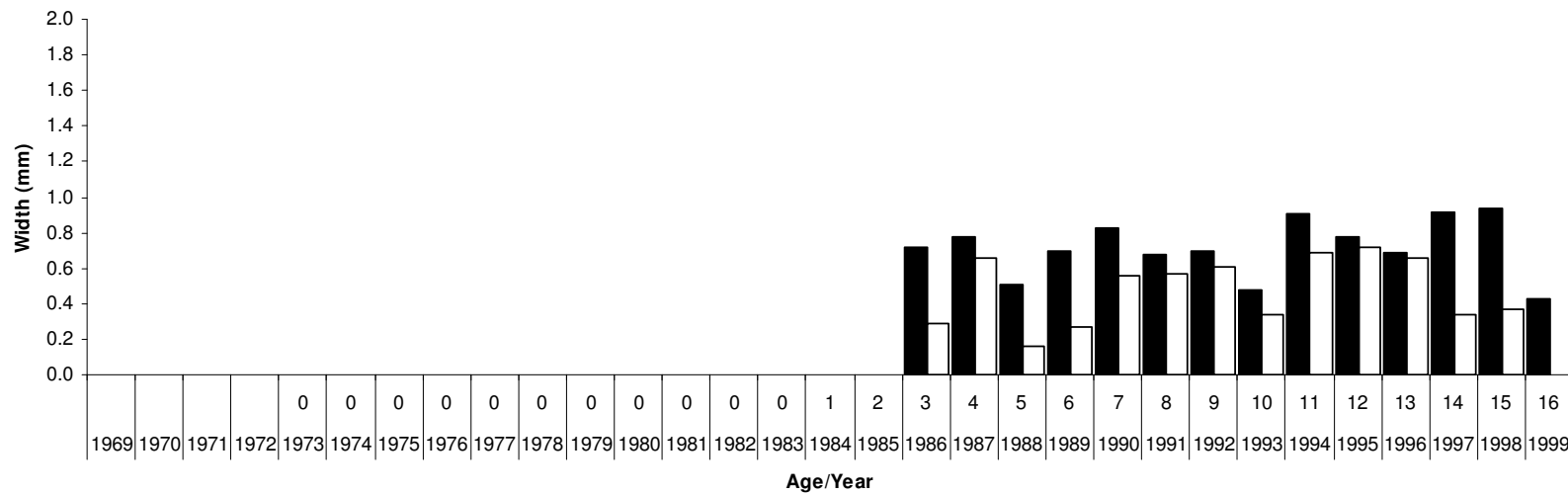
Males:



Appendix Figure 1q. MD21: Male dugong that died in June 1998 at age 16.

Reproductive status: Fully spermatogenic (i.e. mature).

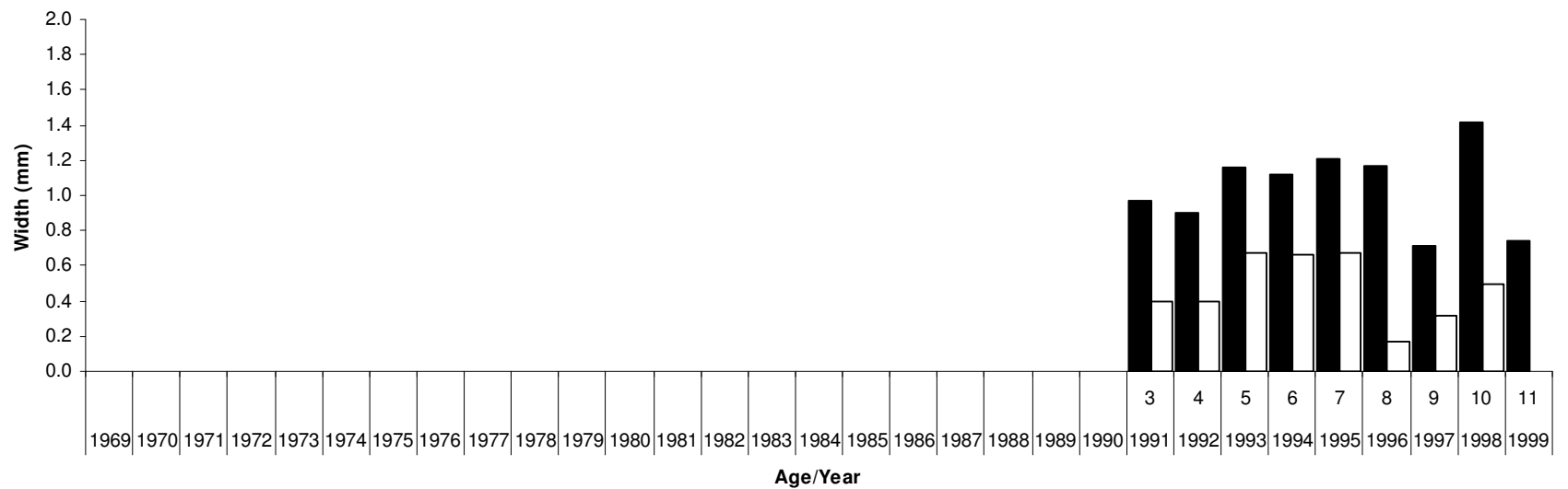
If male dugongs experience a pre-pubertal growth spurt (which is unknown) the tusk record suggests that sexual maturity may have occurred at age 14.



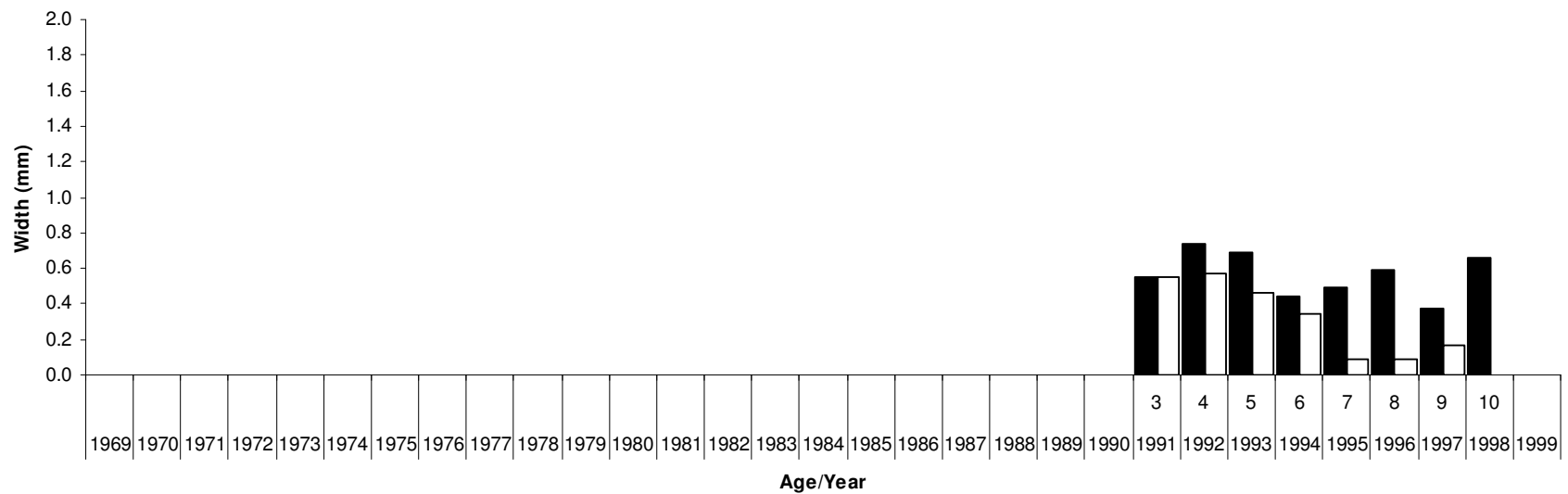
Appendix Figure 1r. MD123: Male dugong that died in April 1999 at age 16.

Reproductive status: Resting (i.e. mature).

If male dugongs experience a pre-pubertal growth spurt (which is unknown) the tusk record suggests that sexual maturity may have occurred at age 14.



Appendix Figure 1s. MD28: Male dugong that died in April 1999 at age 16.
 Reproductive status: Fully spermatogenic (i.e. mature). If male dugongs experience a pre-pubertal growth spurt (which is unknown) the tusk record suggests that sexual maturity may have occurred at age 10.



Appendix Figure 1t. MD26: Male dugong that died in July 1998 at age 10. Reproductive status: Fully spermatogenic (i.e. mature). If male dugongs experience a pre-pubertal growth spurt (which is unknown) the tusk record suggests that sexual maturity may have occurred at age 10.

11. Appendix 2

Calculation of Repeatability

Repeatability, r is given by:

$$r = S^2_A / (S^2 + S^2_A) \quad \text{Equation 1}$$

where, S^2_A is the among-groups (observers) variance component and S^2 is the within-group variance component. The variance components are calculated from the mean squares in the analysis of variance (see Table A2.1) as:

$$S^2 = MS_W \quad \text{Equation 2}$$

and

$$S^2_A = (MS_A - MS_W) / n \quad \text{Equation 3}$$

where n is the sample size per group.

Table A2.1. Analysis of variance for the calculation of repeatability and reliability

Source of variation	Degrees of freedom	Sum of squares	Mean squares	F-ratio
Among groups	df_1	SS_A	MS_A	F
Within groups	df_2	SS_W	MS_W	

As an example the analysis of variance table and calculation of reliability is shown for the case where one observer measures the same 5 GLGs in a tusk each of 4 times, using data from observer 1 and tusk MD56 (Table A2.1).

Table A2.2. Analysis of variance of measurements of widths of GLGs by an individual observer for the ridges of tusk MD56.

Source of variation	Degrees of freedom	Sum of squares	Mean squares	F-ratio
Among times	3	0.021	0.007	0.668
Within times	16	1.637	0.102	
Total	19			

From the analysis of variance table (table A2.2):

$$MS_A = 0.007$$

and

$$MS_W = 0.102$$

Hence, from Equations 2 and 3

$$S^2 = 0.102$$

and

$$S^2_A = (0.007 - 0.102) / 5 = -0.191$$

Substituting into Equation 1,

$$r = -0.191 / (0.102 + -0.191) / 5 = -0.23$$

r is multiplied by 100 to express it as a percentage, -23%.