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Improving the estimation of population risk to Hector's and Māui dolphins (*Cephalorhynchus hectori*) using carcass data, focusing on toxoplasmosis

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Improving the estimation of population risk to Hector's and Māui dolphins (*Cephalorhynchus hectori*) using carcass data, focusing on toxoplasmosis

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Abstract

Toxoplasmosis, a disease caused by the parasite Toxoplasma gondii, for which the domestic cat (Felis catus) is the only definitive host in New Zealand, has been identified as a potentially major cause of mortality in Hector's dolphin (Cephalorhynchus hectori hectori) and Māui dolphin (Cephalorhynchus hectori maui). A spatial multi-threat risk assessment, informing the 2020 Threat Management Plan for both subspecies, used the small available sample of necropsied beachcast dolphins to estimate the population risk from toxoplasmosis. The main potential sources of bias identified were small sample size; spatio-temporal factors, including human, dolphin behavioural, and climatic aspects; longer-term temporal variability in proportional causes of death not represented by the existing necropsy data; factors that may skew the age distribution of the sample; and risk assessment modelling not accounting for a potential female skew in toxoplasmosis deaths. This review includes several recommendations for mitigating or accounting for the identified sources of bias, including the need for an increased carcass detection rate; targeting the winter and spring periods of low beach activity; dedicated effort to find carcasses in locations of high dolphin density but poor public access; and an increased sample size of dead dolphins for necropsy and toxoplasmosis testing. A potential female bias in toxoplasmosis mortalities would pose an increased population risk and could be accounted for by future risk assessment modelling. Future assessments could use fully quantitative methods to estimate the magnitude and direction of the key biases identified here, which could then be included in future risk assessment models fitted to necropsy data.

Keywords: New Zealand, marine mammal, bias, carcass, risk assessment, cetacean

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1. Introduction

1.1 Background

Hector's dolphin (*Cephalorhynchus hectori hectori*) and Māui dolphin (*Cephalorhynchus hectori maui*) are endemic to the coastal waters of New Zealand. At the time of this review, Hector's dolphin was listed as Threatened – Nationally Vulnerable and Māui dolphin as Threatened – Nationally Critical, in the New Zealand Threat Classification System (Baker et al. 2019). At the international level, Hector's and Māui dolphins are listed as Endangered and Critically Endangered, respectively, in the International Union for Conservation of Nature (IUCN) Red List (Reeves et al. 2013a, 2013b). Historically, the conservation of both Hector's dolphin (occurring around the South Island) and Māui dolphin (occurring along the west coast of the North Island) has focused on using area management to mitigate the threat of entanglement and mortality in commercial and recreational fishing nets.¹

More recently, Roe et al. (2013) highlighted the threat to both subspecies posed by toxoplasmosis, a disease caused by the unicellular parasite *Toxoplasma gondii*, based on the findings of formal necropsies of bycaught and beachcast carcasses. In New Zealand, the domestic cat (*Felis catus*) is the only known definitive host for *T. gondii*, shedding millions of oocysts in the weeks after infection, and these oocysts are then capable of infecting all bird and mammal species (reviewed for New Zealand species by Roberts et al. [2020]). The precise pathways by which these dolphins then become infected with *T. gondii* is not known. The infection rates of beachcast individuals examined was approximately 60% in a sample of 28 beachcast or bycaught individuals based on PCR testing of a wide set of tissues (Roe et al. 2013). This rate is high, even compared with many terrestrial species, including humans (Roberts et al. 2020).

A spatial multi-threat risk assessment for Hector's and Māui dolphins (Roberts et al. 2019) was incorporated into the 2020 Threat Management Plan (TMP) for both subspecies (DOC and MPI 2021), using information from the small available sample of necropsied beachcast dolphins to estimate the population risk from mortalities caused by toxoplasmosis. This approach assumed that the causes of death in the beachcast sample were representative of the population. Roberts and Hendriks (2020) identified some factors that could bias this analysis, including a seasonal and spatial concentration in carcass detection rate in months and regions where toxoplasmosis mortality rates are atypical.

Based on the multi-threat risk assessment, the TMP highlighted the need to increase the carcass detection rate and the number of necropsies to better inform the estimation of population risk from toxoplasmosis. However, given the large potential for biasing factors, how could this increased effort be optimised to mitigate or account for biases and improve the assessment of toxoplasmosis risk from carcass data?

The Department of Conservation's (DOC's) draft Toxoplasmosis Science Plan 2021 proposes priority research for filling knowledge gaps to inform management. The first research theme, 'Importance of the risk of toxoplasmosis in Hector's and Māui dolphins', and the priority research criterion 'Improve understanding of the parasite in the dolphins through recovery of dead bodies for necropsy', are directly addressed by this review.

¹ See https://www.mpi.govt.nz/fishing-aquaculture/sustainable-fisheries/managing-the-impact-of-fishing-on-protectedspecies/protecting-hectors-and-maui-dolphins/

1.2 This review

This research builds upon previous work by Roberts and Hendriks (2020), which characterised the information about beachcast Hector's and Māui dolphins and the resulting necropsy data from this sample, focusing on temporal (interannual and monthly) patterns. The purpose of this document is to conduct a more thorough review of the potential causes of bias that may affect the estimation of population risk to Hector's and Māui dolphins from toxoplasmosis and other non-fishery threats when using beachcast carcass necropsy data to make inferences about wider populations.

The specific research objectives of this review were as follows:

- Conduct a review of factors that may bias the estimation of population risk from toxoplasmosis and other causes of death using data from necropsies of beachcast dolphins.
- Specifically with respect to Hector's and Māui dolphins, indicate the probable direction and magnitude of biases for estimating population risk from necropsies of beachcast dolphins, based on the outputs of Objective 1.
- 3. Guided by the outputs of Objectives 1 and 2, provide recommendations as to how efforts to increase carcass detection rates may best be designed to:
 - a. increase the precision of population risk estimates from toxoplasmosis; and
 - b. minimise or account for potential biasing factors for estimating population risk.

The sources of information used in this review include scientific literature, previous analyses on Hector's and Māui dolphins, and other relevant information.

2. Summary of beachcast carcass data

In this section, the reported Hector's and Māui dolphin mortalities are characterised to provide the information requirements for reviewing the potential sources of bias when estimating population risk using the beachcast dolphin sample (see section 3). This characterisation uses only records collated and maintained in DOC's Hector's and Māui Dolphin Incident Database (1921–) ('Hectors-Māuis-incidents-2022-04-28.xls'), which was downloaded from the DOC website on 1 June 2022.

2.1 Temporal patterns

The annual number of Hector's and Māui dolphins reported as beachcast has fluctuated through time, including in the period since 2002, when the sources (e.g., bycatch, beachcast, etc.) of all dolphins in the Hector's and Māui Dolphin Incident Database (1921–) were identified (Fig. 1). Peaks in the number of beachcast individuals reported occurred in 2007–2009, 2012, and 2017–2018. It is not known if this pattern was driven by fluctuations in reporting rate (e.g., these years approximately coincide with the previous TMPs, when the public status of this species will have been relatively high), by changes in the annual mortality rate of the population, or by random chance. Note the relatively low number of beachcast individuals reported annually since 2014 (e.g., relative to the period from 2000 to 2013), as well as the reduced rate of fishery entanglement-related mortalities in both the bycatch and beachcast samples after 2008, when wide-scale fishing area restrictions were implemented around the North and South Islands (Figs 1 and 2) (DOC 2022).

The same data were presented by month to visualise seasonal patterns in the size of the reported beachcast sample since 1985 (Appendix, Fig. A1.1). This reveals a relatively higher rate of reported individuals in the period from October to March, with the fewest samples in winter months. For some causes of death (e.g., entanglement in fishing gear as well as maternal separation), this can largely be explained by known or probable seasonality in deaths (e.g., commercial set net effort peaks in summer in many regions of New Zealand). However, for all causes of death, this also appears to be explained by a seasonal pattern in reporting rate, based on surveys of the number of people visiting beaches throughout the year (the 'beachgoer index' was derived from surf life saving survey data collected from a selection of North Island beaches) (see Roberts and Hendriks 2020).

Relative to the beachgoer index, the number of reported beachcast dolphins was greatest in the period from June to December (Appendix, Fig. A1.1). This may be indicative of a relatively high mortality rate during this period, noting, however, that the seasonal beachgoer patterns may have differed around the South Island, where most of the beachcast sample was located. The same plot is shown disaggregated by sex in Fig. 3. Note the much greater number of beachcast females reported in late winter and early spring, with a peak in October, which was not present for males. Also, a relatively high number of females was reported in September, despite low beach activity in this month. Previously, the proportion of females in the sexed beachcast sample from August to October (0.84) was determined by Roberts and Hendriks (2020) to be significantly different from 0.5 (exact binomial test, P < 0.001, 2-sided). This was not evident in years prior to 2000, although the sexed sample was smaller in these years.



Figure 1. Number of Hector's and Māui dolphins (*Cephalorhynchus hectori*) recovered since 1985 by source, summarised from the 'Observation type' field in the Hector's and Māui Dolphin Incident Database (1921–). Note that 'beachcast' dolphins includes several dolphins that were subsequently determined from necropsies to have died as a result of bycatch (see Fig. 2).



Figure 2. Annual number of Hector's and Māui dolphins (*Cephalorhynchus hectori*) in the beachcast sample since 1985 by primary cause of death determined from necropsy. Summarised from the 'necropsy results' and 'necropsy results details' fields in the Hector's and Māui Dolphin Incident Database (1921–). Here, 'entanglement' included individuals that were identified from necropsy as 'known', 'probable', or 'possible bycatch'. Note that temporal patterns in these data will largely have resulted from changes in necropsy methods through time, particularly for non-fishery causes of death, including toxoplasmosis.

All eight of the beachcast carcasses determined to have died from toxoplasmosis were recovered in the period from September to November (Appendix, Fig. A1.2) (in addition to a Māui dolphin found floating at sea in November).² Based on the increased use of beaches in summer and early autumn (December to March), and consequent higher carcass detection rates, it seems likely that the low detection rates of toxoplasmosis mortality reflect a much lower actual toxoplasmosis mortality rate during these months. Conversely, the much lower use of beaches in winter months means that potential toxoplasmosis mortalities in this period are less likely to be detected.



Figure 3. Monthly number of Hector's and Māui dolphins (*Cephalorhynchus hectori*) in the beachcast sample since 1985 by sex and primary cause of death determined from necropsy. Summarised from the 'necropsy results' and 'necropsy results details' fields in the Hector's and Māui Dolphin Incident Database (1921–). Here, 'entanglement' included individuals that were identified from necropsy as 'known', 'probable', or 'possible bycatch'. Note that comparability of the number of dolphins in each cause of death category, including toxoplasmosis, will be confounded with changes in necropsy methods through time. A 'beachgoer index' is superimposed (black line and points) on the bar plot and gives an approximate indication of the relative number of people on beaches throughout the year (from Roberts and Hendriks 2020).

² Note that a beachcast male Māui dolphin recovered in March 2023, subsequent to the sampling period used by this study, was determined to have died from toxoplasmosis (Wendi D Roe, personal communication, Massey University, Palmerston North, 2023 Apr 21).

2.2 Spatial patterns

Based on an approximate estimate of the annual number of dolphins dying in each region of New Zealand, the proportion of all deaths that were reported as beachcast carcasses (from 2007 to 2021, when necropsy methods have been most consistent) ranged from approximately 0.5% (east coast of the South Island, where the most carcasses were reported) to approximately 11% (west coast of the North Island, which will have primarily comprised Māui dolphins) (Table 1). Given the relatively small total sample of beachcast individuals across all regions in this period (n = 163), the regional distribution of these samples is good, in that, while most are reported along the west and east coasts of the South Island (where the species is most abundant), there is a higher inferred recovery rate of carcasses in smaller populations (such as the west coast of the North Island, where Māui dolphin occurs), which are often prioritised for conservation management.

Table 1. Summary of Hector's and Māui dolphin (*Cephalorhynchus hectori*) data by region, including numbers of dolphins that were beachcast, numbers that were necropsied, and numbers for which toxoplasmosis was identified as the primary cause of death. Also shown are the assumed population sizes for each region and the approximate number of annual deaths that would occur in the average year. For quantities relating to the beachcast sample, these are also expressed as percentages of the total annual number of deaths for each region (across 15 years from 2007 to 2021).

QUANTITY	WCNI	NCSI	ECSI	SCSI	WCSI	REFERENCES
Population size ^a	65 ^b	214	9,728	332	5,482	See table 2 of Roberts et al. (2019)
Approximate annual deaths ^c	6.5	21.4	972.8	33.2	548.2	
Number of beachcast carcasses from 2007 to 2021:						Hector's and Māui Dolphin Incident
all reported	11 (11.28%)	6 (1.87%)	76 (0.52%)	11 (2.21%)	59 (0.72%)	(1921–)
subjected to formal necropsy	9 (9.23%)	5 (1.56%)	55 (0.38%)	10 (2.01%)	44 (0.54%)	
of which a primary cause of death could be identified	6 (6.15%)	3 (0.93%)	37 (0.25%)	5 (1.00%)	22 (0.27%)	
of which toxoplasmosis was identified as the primary cause of death	1 (1.03%)	0 (0.00%)	5 (0.03%)	0 (0.00%)	2 (0.02%)	

Abbreviations: WCNI = west coast of the North Island, NCSI = north coast of the South Island, ECSI = east coast of the South Island, WCSI = west coast of the South Island.

^a Values used for population size were based on rescaling strata used by Roberts et al. (2019).

^b The 2015–2016 population estimate of Māui dolphin (*Cephalorhynchus hectori maui*) was used (63 individuals), which was near the middle of the time series of beachcast dolphins used in this table. This includes two Hector's dolphin (*Cephalorhynchus hectori hectori*) migrants from the South Island that were identified from genetic analysis of biopsy samples (Constantine et al. 2021).

^c Calculated as the total population size multiplied by 0.1 (approximating the annual proportion of dolphins dying each year).

The spatial distribution of beachcast carcasses by primary cause of death is plotted in Figs A1.3 and A1.4 in the Appendix. Overall, the spatial distribution of reported beachcast dolphins is consistent with the estimated at-sea spatial distribution around the South Island and off the west coast of the North Island. There were no obvious spatial patterns when causes of death were compared separately, nor did they show any spatial patterns when compared between regional populations. As summarised in Table 1, dolphins that had died of toxoplasmosis were found beachcast along the east and west coasts of the South Island, as well as one Māui dolphin off the west coast of the North Island. Notably, three of the dolphins that had died of toxoplasmosis were recovered in a region of low estimated at-sea density along the Otago coast, although the sample size is too small to allow for any strong inferences.

In Figs A1.5 to A1.9, Appendix, the fine-scale summer/winter distribution of reported beachcast carcasses is shown relative to the estimated summer/winter at-sea distribution of Hector's and Māui dolphins, the locations of roads, and human population density. The purpose of these plots is to identify potential locations for low carcass reporting rates and to explore some of the human causes of this. For example, the relative lack of carcasses reported south of Raglan (Appendix, Fig. A1.5) may relate to a relatively low at-sea density of dolphins, but also could be linked to the fact that there are fewer coastal access points. Public access is also a strong candidate factor influencing the relatively small number of carcasses from the southern portion of the west coast of the South Island (Appendix, Fig. A1.9).

Furthermore, these plots illustrate how a reduced carcass recovery rate in winter may be related to the more offshore distribution of these dolphins in winter relative to summer (e.g., see Appendix, Fig. A1.7), which could plausibly reduce the likelihood of carcasses washing up along the coastline in this season.

In Figs A1.5 to A1.9, Appendix, the locations of New Zealand rivers are also shown relative to recovered carcasses.

2.3 Demographic composition

Roberts et al. (2019) reported that seven of the nine recorded deaths of Hector's and Māui dolphins were females, of which six were deemed to be reproductively mature based on inspection of reproductive organs. Although the sample size is small, this may indicate a relatively higher propensity of females to die from toxoplasmosis once infected, compared with males. Four of the cases of disseminated toxoplasmosis were aged from tooth sections, including three females (estimated to be ages 9, 14, and 15) and one male (estimated to be age 4) (Roberts et al. 2019).

3. Factors biasing the estimation of population risk

This section reviews the identified factors that may bias the estimation of population risk from toxoplasmosis and other causes of death using data from necropsies of beachcast dolphins. The selected factors are based on the characterisation in section 2, as well as the findings of previous relevant research.

3.1 Small sample size

The relatively small sample size of beachcast Hector's and Māui dolphins, and even smaller sample of dolphins for which a primary cause of death was stated (Table 1), increases the likelihood due to stochasticity that the causes of death in the beachcast sample were not representative of wider patterns. The spatial risk model of Roberts et al. (2019) used a Bayesian implementation of a Dirichlet-multinomial model to generate prior distributions of the proportional non-fishery causes of death, including from toxoplasmosis. This approach would naturally result in increasing uncertainty around the number of toxoplasmosis deaths with decreasing overall sample size, although the mean values for the proportion of deaths caused by toxoplasmosis would still be sensitive to changes in the necropsy proportions, which are more likely at small sample sizes.

The inferred carcass detection rates of Hector's and Māui dolphins varied by region (Table 1), with approximately an order of magnitude greater recovery rate along the west coast of the North Island relative to the South Island regions. This suggests that there is considerable potential to increase carcass detection rates, particularly of Hector's dolphin on the South Island. However, even the carcass detection rate along the west coast of the North Island (approximately 11%) (Table 1) is low relative to that of some other coastal delphinids, e.g., the California coastal population of bottlenose dolphin (*Tursiops truncatus*) (25%; 95% confidence interval = 0.20–0.33) (Carretta et al. 2016) and the Sarasota Bay, Florida, population of the same species (33% of all known deaths and disappearances; standard deviation = 17%) (Wells et al. 2015), although, admittedly, these regions are more densely populated than most coastal regions of New Zealand. Overall, this suggests that population density and the likelihood of people using beaches is likely to have a major effect on carcass detection rate.

3.2 Spatial-seasonal factors

The data characterisation in section 2 highlighted some processes that have probably contributed to the strong seasonal pattern in beachcast carcass recoveries, including highly seasonal beach use and seasonal onshore/offshore movements of Hector's and Māui dolphins. To this list could be added seasonal patterns in weather, sea conditions, and currents affecting the deposition and retention of dolphins on beaches, as well as shifting sands affecting burial/re-exposure, and potentially seasonality in scavenging rate by known marine predators. Of these, only the carcass recovery rate can be directly controlled by interventions. This should ideally increase the carcass detection rate in the winter period, which is largely under-represented in the current sample. This is currently likely to be a biasing factor for the estimation of risk because of the seasonality in toxoplasmosis deaths and other non-fisheries causes of death.

The purpose of the plots in Figs A1.5 to A1.9 (Appendix), is to explore a potential bias that may arise from better public access near river mouths, where Hector's and Māui dolphins may be more likely to be infected with *Toxoplasma gondii* oocysts. This does reveal a tendency for carcasses to be recovered near river mouths in some regions (e.g., Appendix, Figs A1.7 and A1.8), but not all (e.g., Appendix, Figs A1.5 and A1.6). However, since *T. gondii* infection rates are high in the species (approximately 60%, although potentially increasing with age) (Roe et al. 2013, 2015), carcasses were found to be infected with *T. gondii* in nearly all months of the year when they were sampled (Roe et al. 2013), and because there would be a delay between infection and death from toxoplasmosis, it would seem unlikely that toxoplasmosis would be any more likely to kill dolphins near river mouths.

3.3 Long-term changes in causes of death

Necropsy methods used for Hector's and Māui dolphins have been consistent since 2007, although this only provides approximately 15 years of relatively complete information at the time of this review. It is not implausible that the underlying combination of causes of death in the population has changed though time, although we lack a sufficiently long time series of data to assess this. Specifically, with respect to toxoplasmosis, tissues collected during necropsies from 2002 to 2006 were retrospectively assessed for the prevalence of *Toxoplasma gondii* infection, noting that infection is less likely to be detected in older tissues when using PCR testing. Disseminated toxoplasmosis was not detected in any of these carcasses, and should have been detectible in carcasses that were comparatively fresh and on which full histology was done (Wendi D Roe, personal communication, Massey University, Palmerston North, 2022 Jun 15). The significant female skew in beachcast dolphins during August to October (around the time that all toxoplasmosis mortality cases were found beachcast) highlighted by Roberts and Hendriks (2020) was evident for the 2000s and 2010s, but not in the preceding decades, although the sample of beachcast dolphins was much smaller then (Fig. 2).

Taken together, there is some tentative evidence for changes in *T. gondii* infection and disseminated toxoplasmosis through time, which, for example, could cause a positive bias in the estimation of population risk, if the period of deaths was only temporary. However, the sampling period for the data currently available is too short to enable formal assessment of this.

3.4 Demographic composition

Seven of the nine recorded deaths of Hector's and Māui dolphins were females, of which six were adults. An earlier study by Roe et al. (2013) highlighted the presence of *Toxoplasma gondii* in the uterine tissues of multiple necropsied Hector's and Māui dolphins and a case of fatal toxoplasmosis in a pregnant female that had *T. gondii* in foetal, uterine, and placental tissues, and deduced that toxoplasmosis may be an important cause of neonatal deaths. However, Roe et al. (2015) found that males were more likely to be infected with *T. gondii*. Although the sample size is small, this may indicate a propensity for females to die from toxoplasmosis once infected.

The potential skew towards toxoplasmosis fatalities in adult females and the increased likelihood of *T. gondii* infection with age (which is also observed in terrestrial mammals) means that this threat may be under-represented in the beachcast sample if for any reason it was skewed towards younger individuals. For this reason, the use of bycaught dolphins to make inferences about the prevalence of *T. gondii* infection in Hector's and Māui dolphins would be biased if the much higher vulnerability of subadults to being captured

(Davies et al. 2008) was not accounted for. Of those that were aged (*n* = 19), the mean estimated age of the beachcast dolphin sample analysed by Roberts et al. (2019) was 9.0 years. This is slightly above the age at first maturity for this species estimated by Edwards et al. (2018) (6.91 years; 95% credible interval = 5.82–8.24 years), indicating that this sample comprises a representative cross section of the age distribution of the population. This would not be consistent with the beachcast sample being too young or too old to give a representative proportion of deaths from toxoplasmosis, or infections with *T. gondii*.

3.5 Risk modelling approach

The risk assessment of Roberts et al. (2019) was a combined sex model, which did not use existing information that indicated a possible female skew in annual toxoplasmosis deaths. If this skew was representative, excluding this information would have negatively biased the estimation of population risk, on the basis that female Hector's and Māui dolphins are polyandrous breeders, and reproductive output should scale to the number of females of breeding age. Other spatial risk assessments of this nature for other marine mammals have used female-only models (e.g., Large et al. 2019), which may be applicable for this species also, although this would further reduce the sample used for model fitting, resulting in less precise model estimates. This situation would be improved with increased carcass detection rates.

The spatial risk model assessment of Roberts et al. (2019) produced very different estimates of the relative risk posed by commercial fisheries, disease, and other threats, relative to the previous TMP risk assessment, which was based on expert opinion (Currey et al. 2012). Other model-based risk assessments of cetaceans fitted to necropsy data are rare, and have produced conflicting information that may imply inherent biases in beachcast dolphin data, or in the comparative data sources (e.g., Moore and Read 2008). However, this does not mean that the risk assessment approach of Roberts et al. (2019) should itself have biased the mean estimates of annual toxoplasmosis mortalities, which were instead dictated by the proportional causes of death from necropsy data.

4. Discussion

4.1 Limitations of this review

The determination of representative mortality rates of all causes is problematic in cetaceans, given that most deaths occur at sea, and the detection of non-fishery deaths is usually dependent on a carcass washing up on shore, being reported and subsequently observed, and being in a suitable state of preservation for reliable analysis. However, necropsy data can be used to identify the presence of certain threats and can provide information on the relative importance of detectible threats, particularly if biases are identified and accounted for.

This review identified several potential biases, although others may not have been identified. Furthermore, the evaluation of biases was predominantly based on a qualitative and semi-quantitative review of the information. Future research could include fully quantitative/spatio-temporal measurement of the direction and magnitude of the main sources of bias identified here, which could ultimately be included in future statistical risk models that are fitted to necropsy data.

The potential effects of necropsy approaches were not considered in this review, although these are likely to be influential regarding the resulting attributed primary causes of death.

4.2 Probable sources of biases for estimating risk

Based on the consideration of candidate sources of bias in section 3, the probable sources of bias affecting the estimation of toxoplasmosis risk when using beachcast necropsy proportions are summarised in Table 2. Note that some of these sources were listed despite the potential direction of bias (based on existing necropsy data) associated with that source (e.g., small population size, and potential long-term changes in the prevalence of toxoplasmosis) being unknown. For all sources, this summary was based on a qualitative or semi-quantitative exploration of the data and supporting information, whereas fully quantitative approaches would be needed to formally estimate the magnitude and direction of bias.

PROBABLE SOURCE OF BIAS	PROBABLE DIRECTION OF BIAS	PROBABLE MAGNITUDE OF BIAS
Small population size	Neutral, though may depend on risk assessment approach	Increasing probability of bias with decreasing sample size
Seasonality in carcasses being beachcast and reported	Negative, since all toxoplasmosis deaths in the assessed beachcast sample were in spring months when beach activity is low to moderate	Moderate
Spatial distribution of carcass detection rate	Positive, based on beachcast carcasses being predominantly found near river mouths in some, but not all, regions	Moderate in some regions
Sex skew in toxoplasmosis deaths	Negative, since most confirmed cases to date were adult females, although dependent on risk assessment approach used	Strong, if the current sex skew is representative
Long-term changes in the proportion of mortality from toxoplasmosis	Unknown, given lack of longer-term monitoring required to assess this	Unknown, though potentially large

Table 2. Summary of probable sources of bias for the estimation of population risk of toxoplasmosis to Hector's and Māui dolphins (*Cephalorhynchus hectori*). In this table, a positive bias would indicate that the population risk from toxoplasmosis would be over-estimated.

4.3 Conclusions and recommendations

Small sample size issues would be addressed by implementing measures to increase beachcast carcass recovery rates. Encouragingly, while carcass recovery rates are low for this species, particularly around the South Island, a brief spatial exploration of these data found that the regional distribution of carcass recoveries, which to date have primarily been based on public reporting, was good (Table 1). At a finer scale, probable spatial gaps in carcass detection rate appear to primarily be the result of poor public access, indicating that more carcasses may be detected by dedicated (non-public) effort in locations with poor access and high dolphin density. The current approach also appears to result in a representative age distribution in the beachcast sample, whereas, for example, the skew towards subadults in fishery bycatch carcasses (Davies et al. 2008) would be problematic for monitoring *Toxoplasma gondii* infection rates.

The strong seasonal pattern in reported beachcast dolphins was reported previously by Roberts and Hendriks (2020) and appears to be caused by a combination of seasonal beach activity, seasonal movements of the dolphins, and probably also seasonal sea conditions. This seasonality is a potentially major source of bias when using the beachcast sample to make inferences about the population, particularly given the discrete seasonality in deaths primarily attributed to toxoplasmosis, all in the period from September to November, i.e., on the shoulder of the winter period of very low beach activity. Measures to increase carcass detection probability in the winter and spring periods are recommended.

To avoid biasing risk assessments, there is a need to account for the possible skew towards adult female deaths, which may result from the higher vulnerability of females to death from toxoplasmosis. To address this, the carcass recovery rate of females needs to be increased. This could be achieved by not only increasing carcass detection rates in general but also by exploiting the apparent strong female bias in beachcast carcasses detected in late winter and early spring, which was attributed to multiple non-fishery causes of death, including toxoplasmosis.

Finally, future research on sources of bias for toxoplasmosis and other threats could include fully quantitative/spatio-temporal estimation of the direction and magnitude of the main sources of bias identified here, which could ultimately be included in future risk assessment models fitted to necropsy data.

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Appendix

Supplementary data plots



Figure A1.1. Monthly number of Hector's and Māui dolphins (*Cephalorhynchus hectori*) in the beachcast sample since 1985 by primary cause of death determined from necropsy, summarised from the 'necropsy results' and 'necropsy results details' fields in the Hector's and Māui Dolphin Incident Database (1921–). Here, 'entanglement' included individuals that were identified from necropsy as 'known', 'probable', or 'possible bycatch'. Note that comparability of the number of different causes of death will be confounded with changes in necropsy methods through time. A 'beachgoer index' is superimposed (black line and points) on the bar plot and gives an approximate indication of the relative number of people on beaches throughout the year (from Roberts and Hendriks 2020).



Figure A1.2. Monthly number of Hector's and Māui dolphins (*Cephalorhynchus hectori*) in the beachcast sample since 1985 by primary cause of death determined from necropsy, summarised from the 'necropsy results' and 'necropsy results details' fields in the Hector's and Māui Dolphin Incident Database (1921–). Here, 'entanglement' included individuals that were identified from necropsy as 'known', 'probable', or 'possible bycatch'. Note that comparability of the number of different causes of death will be confounded with changes in necropsy methods through time. A 'beachgoer index' is superimposed (black line and points) on each bar plot and gives an approximate indication of the relative number of people on beaches throughout the year (from Roberts and Hendriks 2020).

Beachcast by cause of death



Figure A1.3. Spatial distribution of reported Hector's and Māui dolphin (*Cephalorhynchus hectori*) beachcast events since 1985 along the west coast of the North Island by primary cause of death determined from necropsy. The estimated relative at-sea density of dolphins is also shown (yellow shading = high density, blue shading = low density). Note that a second toxoplasmosis death was identified for an individual found floating 500 m offshore of Raglan that does not appear in the beachcast sample shown here (located approximately 2 km from the beachcast toxoplasmosis death in the plot above).



Figure A1.4. Spatial distribution of reported Hector's and Māui dolphin (*Cephalorhynchus hectori*) beachcast events since 1985 around the South Island by primary cause of death determined from necropsy. The estimated relative at-sea density of dolphins is also shown (yellow shading = high density, blue shading = low density).



Figure A1.5. Spatial distribution of reported Hector's and Māui dolphin (*Cephalorhynchus hectori*) beachcast events since 1985 along the west coast of the North Island in summer (A), winter (B), and all seasons (C). The estimated relative at-sea density of dolphins was square-root transformed and is shown for each respective season, with the summer density shown for the all-seasons plot (yellow shading = high density, blue shading = low density). Public roads are represented by orange lines. Areas of very low human population density appear as black in the left-hand and centre plots. Roads are omitted from the right-hand plot to show the distribution of beachcast dolphins relative to river mouths.



Figure A1.6. Spatial distribution of reported Hector's and Māui dolphin (*Cephalorhynchus hectori*) beachcast events since 1985 along the north coast of the South Island and Kaikōura coast in summer (A), winter (B), and all seasons (C). The estimated relative at-sea density of dolphins was square-root transformed and is shown for each respective season, with the summer density shown for the all-seasons plot (yellow shading = high density, blue shading = low density). Public roads are represented by orange lines. Areas of very low human population density appear as black in the left-hand and centre plots. Roads are omitted from the right-hand plot to show the distribution of beachcast dolphins relative to river mouths.



Figure A1.7. Spatial distribution of reported Hector's and Māui dolphin (*Cephalorhynchus hectori*) beachcast events since 1985 along the Canterbury coast in summer (A), winter (B), and all seasons (C). The estimated relative at-sea density of dolphins was square-root transformed and is shown for each respective season, with the summer density shown for the all-seasons plot (yellow shading = high density, blue shading = low density). Public roads are represented by orange lines. Areas of very low human population density appear as black (in plots A and B only). Roads are omitted from plot C to show the distribution of beachcast dolphins relative to river mouths.



Figure A1.8. Spatial distribution of reported Hector's and Māui dolphin (*Cephalorhynchus hectori*) beachcast events since 1985 along the Otago coast and the south coast of the South Island in summer (A), winter (B), and all seasons (C). The estimated relative at-sea density of dolphins was square-root transformed and is shown for each respective season, with the summer density shown for the all-seasons plot (yellow shading = high density, blue shading = low density). Public roads are represented by orange lines. Areas of very low human population density appear as black (in plots A and B only). Roads are omitted from plot C to show the distribution of beachcast dolphins relative to river mouths.



Figure A1.9. Spatial distribution of reported Hector's and Māui dolphin (*Cephalorhynchus hectori*) beachcast events since 1985 along the west coast of the South Island in summer (A), winter (B), and all seasons (C). The estimated relative at-sea density of dolphins was square-root transformed and is shown for each respective season, with the summer density shown for the all-seasons plot (yellow shading = high density, blue shading = low density). Public roads are represented by orange lines. Areas of very low human population density appear as black in plots A and B. Roads are omitted from plot C to show the distribution of beachcast dolphins relative to river mouths.