An analysis of historical bottlenose dolphin (*Tursiops truncatus*) strandings in the Mississippi Sound, USA using classification and regression trees (CART)

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ABSTRACT

Trends in bottlenose dolphin (*Tursiops truncatus*) strandings can be used to examine several factors associated with mortality and life history and are essential for detecting unusual mortality events (UMEs). This study characterised stranding trends in the Mississippi Sound (MS) region of the northern Gulf of Mexico (GoM) from 1996–2009 using kernel density estimation (KDE) and classification and regression tree (CART) analysis. An annual mean of 26.1 strandings (*n* = 14), SD = 13.7, 95% CI [18.2, 34.0] and a peak in strandings during spring (March–May) were evident from our analyses. Neonates stranded almost exclusively in spring indicating that this is the dominant breeding and calving season in this area. Spatial distributions revealed that the majority of dolphins stranded along central and western portions of the MS Sound near Gulfport, MS and on Ship Island during the spring and summer months, but were more often found in the eastern MS Sound during winter and autumn. Our CART analyses indicated that 1996, which contained a declared UME, was anomalous from other years as the number of adult, sub-adult and juvenile strandings was relatively high during the autumn and winter. Further, our analyses showed that the location of those strandings on Ship Island in autumn and winter was unique from all other years in the historical record. These results represent historical conditions that can be used as a baseline for future studies of the effects of environmental disturbances, including UMEs, in MS. This research also demonstrates the versatility and usefulness of CART for describing historical trends, detecting departures from the norm and explaining UMEs within the framework of a single analysis. This approach represents an objective assessment tool that could be used to assist governmental agencies with determining the onset of a UME and could help support or refute the cause of these events.

KEYWORDS: COMMON BOTTLENOSE DOLPHIN; STRANDINGS; GULF OF MEXICO; NORTHERN HEMISPHERE; STATISTICS; DISTRIBUTION

INTRODUCTION

The enactment of the US Marine Mammal Protection Act (MPA) in 1972 and amendments in 1992 that created the Marine Mammal Stranding Network have generated increased awareness of bottlenose dolphin (*Tursiops truncates*) strandings in the USA (McFee and Hopkins-Murphy, 2001). Since that time, much has been learned about the species as strandings have provided unique opportunities to study factors associated with mortality (Lipscomb et al., 1996; Meador et al., 1999) and life history (Hubard and Swartz, 2000; Mattson et al., 2006; McFee and Hopkins-Murphy, 2001; McFee et al., 2006) that have influenced management and conservation of this protected species. Further, because bottlenose dolphins are considered a sentinel species (Wells et al., 2004) periodic assessment of stranding trends is critical for gauging the effects of environmental perturbations on ecosystem health.

A major focus of strandings research is Unusual Mortality Events (UMEs), which can result from viral infection (Lipscomb et al., 1996), bacterial infection (McFee and Lipscomb, 2009), biotoxins (MMC, 1996) and many other potential factors. A UME is defined in the MMPA as ‘a stranding that is unexpected; involves a significant die-off of any marine mammal population; and demands immediate response.’ The Working Group on Marine Mammal UMEs, composed of members from scientific and academic institutions, conservation organisations and federal and state agencies, is charged with examining the nature of a stranding event and determines if the event should be declared a UME. Criteria developed by the working group (and see Gulland, 2006) for determining the onset of a UME are:

1. unusual magnitude;
2. marked change in temporal dynamics;
3. marked change in spatial dynamics;
4. marked change in species, age, or sex distributions;
5. unusual pathologic findings, behaviour patterns, clinical signs, or physical condition;
6. mortality or morbidity among depleted, threatened, or endangered populations; and
7. stranding of critically endangered species.

These criteria, while necessary, are somewhat subjective as no formal quantitative method for determining the onset of a UME is defined. With respect to the unusual magnitude of a stranding event (Criteria 1), National Marine Fisheries Service (NMFS) has stated ‘There is no set formula for determining what magnitude would trigger a response. The NMFS Southwest region has used a formula of the historic mean plus two times the standard deviation to determine a threshold level,’ but also states that the magnitude of a stranding event must be weighed against other knowledge (NOAA, 2004). While there is no replacement for expertise of the working group, there is a need for quantitative

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methods that incorporate such varied information into a single analysis. Application of statistical tools that complement the expertise of the Marine Mammal Working Group are essential for assessing the unusual nature of a marine mammal mortality event.

The northern Gulf of Mexico (GoM) is an area with dense populations of bottlenose dolphins (Waring et al., 2009) that has been subject to numerous environmental disturbances over the last several years including freshwater flood events (Carmichael et al., 2012), hurricanes (Miller et al., 2010), and oil spills (Rico-Martinez et al., 2013) that have impacted ecosystem health in the region. Coinciding with these events is the longest running UME on record in the northern GoM. The UME began in 2010 and continues to date, and has included 1,271 cetacean strandings over a region extending from the western border of Louisiana to the Florida Panhandle. While the cause remains unknown, several potential factors may have contributed to this UME including a prolonged bout of cold weather, unusually large freshwater floods (Carmichael et al., 2012), disease and the Deep Water Horizon (DWH) oil spill. While the duration, geographic spread and magnitude of this event have been unique compared to past UMEs, a thorough understanding of historical stranding trends is needed to provide essential context for assessing spatial, temporal or demographic departures from the norm.

In this paper, a historical assessment of bottlenose dolphin strandings is presented for the Mississippi (MS) Sound from 1996–2009, to describe pre-UME stranding trends in this region. Our analysis approach highlights classification and regression tree analyses (CART), which we believe is well suited for identifying patterns in strandings including anomalous events, and thus could be used as a quantitative assessment tool for identifying UMEs.

METHODS

Study Area

The MS Sound encompasses a 2,000 km² area separated from the larger GoM by six barrier islands (i.e. Cat, West Ship, East Ship, Horn, Petit Bios and Dauphin Islands) (Eleuterius, 1978) (Fig. 1). Water depth ranges from 1 to 7m, mean annual water temperature ranges from 9°C in winter to 32°C in summer and salinity ranges from 0 to 33 parts per thousand (ppt) (Christmas, 1973). Population estimates in 2007/08 indicated the total number of dolphins ranges from 2,255 during the summer months to 1,413 during the winter (Miller et al., 2013) indicating that this is among the most densely populated areas within the northern GoM.

Data collection

Data collected for this study were obtained from the NMFS database, which archives data received from state marine mammal stranding networks (MMSN). The primary stranding respondent and member of the MMSN in MS is the Institute for Marine Mammal Studies (IMMS), which has been an active member of the national stranding network since 1984, and currently responds to all strandings in MS. Information collected by respondents for this study included...
to explain stranding distributions including: (1) the tree within one standard deviation of the smallest cross-validation error (CVE) (Breiman et al., 1984; De’ath and Fabricius, 2000); and (2) the smallest tree that explained the most variation in stranding counts (De’ath, 2002). Cross-validation error is a measure of predictive accuracy and is often used for selecting trees to predict unobserved data where a value of zero indicates perfect prediction and values near or above one are indicative of poorer predictive capacity (De’ath, 2002). Classification and regression trees constructed here were not used for prediction; however, relative CVE was used to select the best descriptive trees that contained the most useful information (De’ath and Fabricius, 2000). For regression trees, the proportion of the total sum of squares (SS) explained by the tree was reported. For classification trees, the misclassification rates (MCR) and null model misclassification rates (NMMCR) were reported, which represents a random prediction (De’ath and Fabricius, 2000).

RESULTS

From 1996 to 2009, National Oceanic and Atmospheric Administration level A stranding reports were filed for 366 bottlenose dolphins in the MS Sound (Fig. 1). The spatial distribution of strandings in the MS Sound determined by KDE over the historical record (1996–2009) indicated the highest stranding densities (6–10) occurred along a portion of mainland MS Coast between Biloxi Bay and Bay St. Louis and in the central portion of Ship Island (Fig. 2). Estimates also showed moderate stranding densities (2–6) concentrated throughout central portions of the MS Coast between Biloxi Bay in the east and Bay St. Louis in the west and isolated clusters west of Bay St. Louis, near Ocean Springs, near Pascagoula and on Ship and Horn Islands.

Mean annual strandings during the study period were 26.1 (n = 14), SD = 13.7, 95% CI [18.2, 34.0] and ranged from 8 in 2007 to 59 in 1996 (Fig. 3). Based on these data, an annual stranding level at or above 53.5 would be considered a marked increase in magnitude (UME Criteria 1; Gulland, 2006). The total number of strandings in 1996 exceeded this level and was declared a UME. Mean number of strandings was highest in the spring for all age classes but showed no clear pattern among other seasons (Fig. 4). The mean for the adult age class ranged from 0.92 (n = 14), SD = 1.63, 95%
CI [0, 1.9] in autumn to 1.79 (n = 14), SD = 1.31, 95% CI [1, 2.5] in spring (Fig. 4a). Strandings in the perinate age class varied most widely among seasons as they ranged from 0 (n = 14) in summer to 4.7 (n = 14), SD = 3.1, 95% CI [2.9, 6.5] in spring (Fig. 4d). Adult, sub-adult, and juvenile strandings exceeded the mean ±2SD for each respective age class during autumn of 1996 (Figs 4a, b, and c). Adults and juveniles also exceeded this level in winter of 1996.

A regression tree was constructed to explain variation in stranding counts as a function of year, season and age class. The regression tree was pruned based on the relation between relative and CVE and regression tree size and resulted in selection of two trees to explain the distribution of strandings among selected factors (Fig. 5). The first tree contained the lowest CVE (0.87) and had one division among seasons that explained 18% of the total SS (Fig. 6a). This division divided spring strandings from all other seasons. The mean number of spring strandings for each age class within each year was 2.88 (n = 56), SD = 2.6, 95% CI [2.2, 3.6]. The second regression tree (CVE = 0.92) contained a total of five divisions and explained 44% of the SS, where the length of each branch was related to the relative amount of the SS explained by each division in the tree (Fig. 6b). The tree divided spring strandings by age class (7% of total SS) where perinates, with a mean of 4.7 (n = 14), SD = 3.1, 95% CI [2.9, 6.5] were different than adults, juveniles and sub-adults with a mean of 2.26 (n = 42), SD = 2.2, 95% CI [1.6, 2.9] indicating that perinates had a clearly defined stranding peak in spring relative to the other age classes. The left branch of the tree showed that strandings in winter, summer and autumn were divided by year before or after 1997 (9% of total SS). This indicated that 1996 non-spring strandings were unique from all other years of the study period. The next division was among 1996 strandings, which showed that no perinates stranded in autumn, summer or winter (n = 3) (5% of total SS). The last division showed that adult, juvenile and sub-adult strandings were concentrated in the autumn and winter and with a mean of 6.17 (n = 6), SD = 2.1, 95% CI [3.9, 8.4] compared to a mean of 1.33 (n = 3), SD = 1.5, 95% CI [0, 5.1] for summer strandings (5% of total SS).

A classification tree was constructed to explain the spatial distribution of strandings as a function of year, season and age class. For this analysis, the tree with the smallest CVE (0.99) was also the smallest tree that best explained spatial location of strandings (Fig. 7). This tree had an MCR of 71% (MMNMR = 83%). The first division split strandings from 2009 from all other strandings over the period of record. The terminal leaf of the right branch showed that 2008–2009 strandings occurred predominantly along the central MS coast in Gulfport (12) and Pass Christian (9). The left branch of the tree was further divided by year before or after 1997. Strandings that occurred in 1996 occurred predominantly in Gulfport (14) and on Ship Island (14). Strandings from 1997–2007 were further divided by season, where summer strandings were most commonly found along the central MS coast in Gulfport (9) and Long Beach (8). Fall, winter, and spring season strandings from 1997–2007 were further divided by age class where juveniles, which stranded most
often in Biloxi (13), were different from adult, perinate, and sub-adult strandings, which occurred most often on Horn Island (22) in the eastern MS Sound, and in Gulfport (19) and on Ship Island (18).

DISCUSSION
Temporal and demographic trends
The results show that MS has a mean annual bottlenose dolphin stranding rate of 26.1 and highly varied annual strandings over the 14 year period prior to the onset of the current northern GoM UME. It is difficult to determine the effects of surveillance effort on this estimate; however, the occurrence of Hurricane George in 1998 and Katrina in 2005 may have reduced stranding response activity. This seems more evident during the period from 2005–07, which had very low annual totals that may have resulted from reduced surveillance effort following Hurricane Katrina. Other studies have used temporal blocking to try and account for difference in effort (McLellan et al., 2002), but the inability to effectively quantify effort in MS in this study made a

Fig. 4. Mean number of strandings of known length in the Mississippi Sound from 1996–2006 for (a) adults (>247cm), (b) sub-adults (227–247cm), (c) juveniles (115 – 227cm) and (d) perinates <115cm for each season of the year. Seasons were defined as winter (December–February), spring (March – May), summer (June – August), and autumn (September–November). The sample size for each estimate of the mean is 14, which corresponds to the total number of seasons among years that a stranding total was recorded. Error bars correspond to the standard deviation of the mean.
temporal blocking scheme inadequate to address this deficiency. Further, the regression tree did not detect any differences in stranding abundance for this time period aside from 1996.

Our results indicated that the spring season (March–May) was the dominant peak stranding season for all age classes, especially perinates. The unimodal distribution of neonate strandings suggests that spring is the dominant breeding and
calving season in the MS Sound, which is in agreement with what has been reported for MS (Mattson et al., 2006) and other areas within the northern GoM (Fernandez and Hohn, 1998; Wursig et al., 2000). Seasonal stranding trends among other age classes were less pronounced, but exhibited peaks in spring as well. Interestingly, this contrasts with bimodal distributions of strandings in portions of the Atlantic Coast that exhibit a spring and autumn peak (McFee and Hopkins-Murphy, 2001; McFee et al., 2006). In North and South Carolina, distinct peaks in neonate strandings are seen in autumn and spring (McFee et al., 2006), indicating an autumn and spring breeding and calving season in this area.

The year 1996, which contained a declared UME, had the highest number of strandings and was determined to be unique from all other years with regard to the total number of strandings. Regression trees constructed using temporal and demographic explanatory factors revealed that unusual increases in adult, juvenile and sub-adult strandings occurred in autumn and winter of the year, outside of the typical peak stranding season. Historical reports from the area document that this event contained a total of 31 bottlenose dolphins in November and December that were believed to have stranded as a result of a harmful algal bloom (Karenia brevis) (MMC, 1996).

Spatial trends
Spatial trends revealed from KDE indicated that strandings are most often found along the central and western portions of the MS mainland. Classification trees confirmed these findings and showed that juvenile, perinate and sub-adult strandings were most often found along the central MS Coast in cities such as Gulfport during the spring and summer. Such areas have dense human populations and are regularly visited by residents and tourists, thus surveillance effort is naturally higher here than in more isolated areas such as the barrier islands. Ship Island was an exception as KDE also revealed dense clusters of strandings occurred here; however, Ship Island is regularly visited by tourists via ferry from March–October as weather allows. Thus, surveillance effort cannot be ignored when considering these spatial distributions. Since 2010, governmental surveillance has increased throughout all portions of the MS resulting from the UME investigation and the concurrent Natural Resource Damage Assessment (NRDA) investigation. This must be considered in future studies that compare post-oil spill trends with historical trends, and for identifying future UMEs as varied surveillance effort over time and space makes it difficult to determine if stranding trends represent true departures from the historical norm (McLellan et al., 2002). Cetacean drift must also be considered as strandings can drift long distances after death depending on tides, winds, decomposition state and the size and buoyancy of the animal (McLellan et al., 2002; Peltier et al., 2012). The presence of stranding clusters in central and western portions of the MS Sound could partially be the result of a prevalent southeast to northwest current (Morton, 2008). This would indicate that the majority of strandings originated in the central and eastern portions of the MS Sound.

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**Fig. 7.** Classification trees constructed from bottlenose dolphin strandings in the Mississippi Sound from 1996–2009 showing a tree with four divisions (CVE = 1.01; SE = 0.02). The spatial distribution of strandings is modelled as a function of year, season, and age class. Seasons include spring, summer, autumn, and winter. Age classes include adult (Ad), sub-adult (SA) juvenile (Juv) and perinate (Pe). The length of each branch corresponds directly to the amount of variance explained by the corresponding factor. The distribution of strandings is displayed as a bar graph at each terminal leaf showing the frequency of strandings at each location. Written below each terminal leaf is the location with the largest number of strandings along with sequential totals for each corresponding location in the displayed bar graph.
Also important for interpreting spatial trends are the abundance and distribution of live dolphins. The abundance of dolphins in MS Sound fluctuates considerably over the course of a year with higher densities in summer and lower densities in winter (Hubard et al., 2004; Miller et al., 2013). A large majority of dolphins (73.5%) are considered transients that leave the area during the colder months of the year (Smith et al., 2013). No peer-reviewed research has examined spatial distributions of bottlenose dolphins in the entire MS Sound. Previous population research focused in the eastern MS Sound has determined wide distributions within that area throughout the year (Hubard et al., 2004; Miller et al., 2013). Future studies aimed at examining population dynamics should include greater emphasis on spatial distributions for the entire MS Sound, which will enhance management and protection of dolphins in this region and may provide important clues for understanding stranding trends, including those associated with UMEs.

The cause of death for strandings is impossible to determine with such a large number of factors at play, but theories can be supported or refuted based on the available evidence (Carmicheal et al., 2012; McFee and Lipscomb, 2009). Classification trees in this study revealed unique spatial trends in stranding distributions in 1996. A total of 14 strandings were found on Ship Island in 1996 compared to a total of 28 Ship Island strandings for all other years combined. As mentioned previously, this event is believed to be the result of an algal bloom. Repeated testing on Cat Island (just west of Ship Island) showed that marine dinoflagellate cells (K. brevis) rose from 13,000 cells per liter on November 14 1996 to 13 million cells per liter on November 20 1996 (MMC, 1996) coinciding with the UME. Such large numbers of strandings on Ship Island and the high concentration of K. brevis in that area further support the idea that an algal bloom may have caused this UME and indicate that it may have been most prevalent near the western barrier islands. While the cause of death for these strandings cannot be verified, a clear link between geographic and ecological data is reliable evidence that supports K. brevis as a cause of this event. The ability to link geographic and ecological data may be even more critical for detecting emerging diseases in cetaceans (McFee and Lipscomb, 2009). 

**Application of CART for exploring stranding trends**

This study effectively demonstrated the usefulness of CART for exploring stranding trends. The ability to incorporate different types of data that exhibit varying distributions and missing values into a single analysis (De’ath and Fabricius, 2000; De’ath, 2002) makes CART a versatile tool for exploring stranding data. Historical trends such as peak stranding season and associated demographics were readily defined in our analyses as was the spatial distribution of strandings. More importantly, this approach was able to identify anomalous stranding trends across multiple explanatory factors simultaneously to detect a UME and explain its uniqueness. Factors associated with four of the seven criteria for determining the onset of a UME including those related to magnitude, timing, spatial location and age composition were incorporated. The remaining criteria could also be incorporated if necessary to account for different species of marine mammals, unique pathologies and behaviour patterns. Thus, this approach is a valuable resource to complement the expertise of the Marine Mammal Working Group for determining the onset of a UME within the context of historical data.

CART is an efficient means to explore data and contains easily understandable output that lends itself to explanation (De’ath and Fabricius, 2000; De’ath, 2002). Although strandings in 1996 were more than two SDs above the historical average and were thus part of a declared UME, CART showed that the unusual magnitude was among adult, sub-adult and juvenile age classes out of the typical peak stranding season. Also, CART was able to show that while autumn and winter strandings were typically distributed throughout the MS Coast in autumn and winter, an unusual magnitude of juvenile and sub-adults were stranding on Ship Island in close proximity to an area affected by an algal bloom. Obviously, the declaration of the 1996 UME was made without use of CART; however, it could have helped to efficiently explain the unusual nature of this event and provided greater support for an algal bloom as the cause. Shifting spatial distributions for strandings from 2008–2009 indicate an unusually large number of strandings occurred in Pass Christian, which may not necessarily indicate a UME, but does demonstrate the ability of CART to detect variations in stranding spatial location that could otherwise go unnoticed. Application of a quantitative assessment tool such as CART, that increases objectivity and provides evidence to help to determine the cause of a UME, is important for improved protection of cetaceans worldwide.

The current UME in the northern GoM is an excellent example of an event that has several suspected causes including the DWH oil spill, large freshwater flow events, prolonged bouts of cold weather and the bacterium *Brucella*, among others. Determining the cause and unusual nature of this event relies heavily on well-defined historical trends and adoption of quantitative assessment approaches to assess departures from the norm. Despite the increased awareness and attention cetacean strandings have received in this region, no historical analyses of stranding trends have been published in peer-reviewed literature for MS. Thus, historical stranding trends have not been adequately explored and deserve greater attention as context for understanding the uniqueness of the current UME. Undoubtedly, more work needs to be done to address an incomplete understanding of all aspects of bottlenose dolphin ecology in the northern GoM as this is a critical, yet poorly studied habitat for this sentinel species (Waring et al., 2009).

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