

## The Northern Gulf of Mexico Cetacean UME: Comparisons of UME bottlenose dolphin (*Tursiops truncatus*) strandings with historical data (2000-2011)

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The Northern Gulf of Mexico Cetacean Unusual Mortality Event (UME) began in February of 2010 and is ongoing as of May 2011. The geographic extent of the UME is the Louisiana/Texas border through Franklin County, Florida and includes more than 450 cetaceans, 91% of which are *Tursiops truncatus*. From February through April 2010, stranding rates were elevated in the UME area and included a marked increase in stranding reports around Lake Pontchartrain, LA. Stranding rates continued to be elevated following the Deepwater Horizon oil spill on April 20<sup>th</sup>, 2010. From January – April, 2011, a large proportion of stranded dolphins were premature or perinatal (40%). Age, sex, and geographical distributions of available stranding data from the current UME (February 2010-April 2011) were compared with previous strandings in the Gulf of Mexico (January 2000-January 2010). There were no significant differences in sex distribution between the current UME and baselines. A significantly higher proportion of strandings were reported in Alabama, Louisiana, and Mississippi during the current UME compared to baseline. Stranded perinates ( $\leq 115$ cm) and neonates (116-137 cm) were more prevalent in the current UME compared to overall baselines. In summary, the young age and geographic location of stranded dolphins from the current UME differs from the ten-year baseline. The UME investigation is ongoing and will proceed as outlined in the National Contingency Plan for Response to Unusual Marine Mammal Mortality Events ([http://www.nmfs.noaa.gov/pr/pdfs/health/umme\\_contingency.pdf](http://www.nmfs.noaa.gov/pr/pdfs/health/umme_contingency.pdf)).

## Rapid differentiation of *Kogia sima* and *K. breviceps* by high-resolution melt analysis

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The genus *Kogia* includes two species, *K. sima* and *K. breviceps*. It is difficult to distinguish them due to their morphological similarities. PCR-based identification, including DNA extraction (1 h), PCR (1.5-2 h), electrophoresis (0.5 h) and DNA sequencing (2 d) and following analyses, is the most often technique used for differentiating these

two species. Here we report a quick and efficient method for distinguishing the two species, based on differences in high-resolution melt (HRM) curves of dsDNA. First we quickly extracted DNA from the tissues by using QuickExtract™ DNA Extraction Solution 1.0 (about 15 m), and then used TAQXpedite™ PCR System to rapidly amplify DNA fragments. A pair of primers that target sequences within the highly variable mitochondrial DNA control region was designed. Two-step PCR was carried out as follows: initial denaturation at 98°C for 50 s, followed by 25 cycles of denaturation at 95°C for 10 s, annealing and extension at 65°C for 20 s; and one final extension step at 68°C for 3 m. The PCR product is expected to be approximately 420 bp in size, and a clear single band was visible by electrophoresis in 1.5% agarose gel. Then we performed HRM analysis of the PCR products in the presence of the LCGreen fluorescent dye with the LightScanner 32 instrument, which could be finished in 10 m. *K. sima* and *K. breviceps* generated distinct melt curve profiles, so we used the melting profiles as molecular fingerprints for species typing. For future application, this HRM technique substantially simplifies the work needed to confirm species identification in *Kogia*, especially only small amount of tissue is needed and confirmation can be made with speed.

## Use of Micro-Computed Tomography for dental studies in modern and fossil odontocetes

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Teeth are important elements in studies of modern and fossil cetaceans, providing information on feeding habits, estimations of age, and phylogenetic relationships. The growth layer groups (GLGs) recorded in dentine have demonstrated application for aging studies, but also have the potential to elucidate life history phenomena such as metabolic or physiologic events. Micro-Computed Tomography (Micro-CT) is a non-invasive and non-destructive technique that allows 3-dimensional study of mineralized tissues and their physical properties. It has mostly been used for qualitative dental studies in humans. Teeth from extant dolphins (*Globicephala* sp. and *Sotalia guianensis*) and an unnamed Oligocene fossil dolphin (OU 22108) were scanned in a Skyscan 1172 Micro-CT desktop system. X-rays were generated at 100 kV and 100  $\mu$ A for extant samples, and at 80kV and 124  $\mu$ A for the fossil tooth. 0.5 mm thick aluminum and copper filters were used in the beam. Reconstructed images were finely resolved for the fossil, showing the enamel, internal layers of dentine and the pulp cavity. The enamel layer was well defined in both extant species throughout the images, but the dentinal layers were less resolved. We are refining the use of Micro-CT for dental studies in cetaceans, to allow resolution of internal structure and potential application in non-destructive aging techniques. Imaging software should elucidate greyscale values observed in the dentinal region of extant specimens and their relation to GLGs. Future Micro-CT analysis will involve paired scans of teeth alongside resin-hydroxyapatite calibration standards of known densities to quantify the mineral density of dental tissues in odontocetes.

## Diurnal Behavior and Group Size Patterns of Common Dolphins (*Delphinus* sp.) during 2008-2010 Aerial Surveys off San Diego, California

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Aerial surveys offer an ideal observation platform to document the behavior and group size of dolphin species over a wide range in offshore waters in a short period. Eight aerial surveys were conducted near San Clemente Island off southern California, Oct/Nov 2008, Jun/Jul/Nov 2009 and May/Jul/Sep 2010 to monitor behavior of marine mammal species using line-transect and focal-behavioral circling methods. An estimated 94,867 short-beaked (*Delphinus delphis*) and long-beaked (*D. capensis*) common dolphins were observed during 346 separate events. Number of sightings, mean group size and initial group behavior state were recorded and compared by diurnal periods. Daytime observation hours were divided into three periods, following the methods of Bearzi et al. (1999): “morning” (0801-1159)(n = 71 sightings, individuals = 22,777), “early afternoon” (1200-1559)(n = 191, individuals = 52,926), and “late afternoon” (1600-1959)(n = 85, individuals = 19,164). Sighting rates were highest in the early afternoon (4.1 indiv/km) followed by late afternoon (3.9 indiv/km) and morning (3.3 indiv/km). Mean group size was highest in the “morning” (321 ± 455.8), followed by “early afternoon” (277 ± 354.5) and “late afternoon” (225.5 ± 276.0). During the “morning”, initial group behavior was most frequently surface-active mill (65%) followed by travel(29%). During the “early afternoon”, travel (36%) occurred most frequently, followed equally by surface-active travel (28%) and surface-active mill (28%). During the “late afternoon”, travel (37%) and surface-active mill (36%) occurred most frequently, followed by surface-active travel (27%). Social and apparent foraging behaviors typically occurred during surface-active behavior states. Results suggest that groups of common dolphins of both species aggregate in larger numbers and are generally more surface-active during the morning than early and late afternoon, potentially corresponding with socializing and foraging strategies. Further data gathering and multivariate analyses are underway to elucidate more specific diurnal trends and behavioral correlations.

### Spatial Use Patterns of Ribbon and Spotted Seals in the Bering and Chukchi Seas

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Ribbon and spotted seals rely on the seasonal sea ice of the Bering Sea for pupping and breeding, yet information vital to understanding seasonal changes in the spatial use patterns of these species is scarce. Between 2007 and 2010, 40 ribbon seals and 22 spotted seals were captured in the central and western Bering Sea and released with satellite tags. A continuous-time, correlated random walk, state-space model with importance sampling was used to incorporate location and model uncertainty into a spatial use analysis. This analysis was one of the first to incorporate the newly implemented Argos Kalman filter algorithm into the location error process of a state-space model. Spatial use grids were created for each species across three seasons: March to May (the ice-associated pupping and breeding period), June to October (mostly non-ice associated), and November to February (returning to the newly-formed ice prior to pupping). Tagged individuals from both species were concentrated within the marginal sea-ice zone during pupping and breeding. During the relatively ice-free period, ribbon seals expanded their range north along the Russian coast into the western Chukchi Sea and Arctic Ocean, and as far south and east as the Gulf of Alaska. During this same period, spotted seals remained almost entirely on the Bering and Chukchi shelf. The average portion of time spent north of the Bering Strait during this time was 0.095 (sd: 0.257) for ribbon seals and 0.261 (sd: 0.372) for spotted seals. The range of both species shrank as ice coverage increased between November and February. This study provides important insight into the seasonal spatial-use patterns of ribbon and

spotted seals and how these species may be impacted by future reduced sea-ice coverage.

### Using spatial analysis techniques to identify clustering patterns of North Atlantic right whales in Jeffreys Ledge and investigate associations between sightings and bathymetric features

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Jeffreys Ledge, located off of Cape Ann, Massachusetts, is not generally considered a major habitat for the North Atlantic right whale; however, recent studies have documented that right whale sightings occur there predictably between October and December. The identification of up to 50 individuals within a given sighting season suggests that Jeffreys Ledge may be a secondary habitat for right whales. This study examines where clustering occurs around Jeffreys Ledge and whether sightings are associated with two bathymetric features: depth and degree of slope. Between 2003 and 2009 the Whale Center of New England conducted boat-based surveys for right whales between 15 September and 30 December following systematic track lines along ledge contours. Sightings were corrected for daily trackline effort and entered into ArcGIS. The region was divided into 2 km<sup>2</sup> grid cells and the study area was defined as all grid cells within 5 km of the tracklines. Data were aggregated by cell such that each cell contained the sum of the SPUE from individual surveys, the average depth, and the average slope. To locate areas of whale clustering, the Hot Spot Analysis (Getis-Ord Gi\*) tool in ArcGIS was used. The analysis identifies a hot spot restricted to the northwestern slope of the ledge which changed in size and location between 2003 and 2009. To determine whether bathymetric features influenced the probability of right whale presence, a binary logistic regression was performed using all grid cells in the study area. Results indicate that the probability of seeing a whale increases significantly with depth, but not slope. To determine whether the number of whales seen varied by slope or depth, a regression was performed using aggregated SPUE, only on cells containing whale sightings; SPUE in a grid cell increases with average depth, but slope has no effect.

### Feeding habits of Guiana dolphins, *Sotalia guianensis*, in Brazilian waters

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The knowledge on the feeding habits of Guiana dolphins comes mainly from studies based on the analysis of stomach contents. A total of 341 stomach contents were analyzed in 16 studies conducted in Brazilian waters from 1963 to 2011. To verify prey importance, 50.0% of these studies used the Index of Relative Importance (IRI) as main methodology, 25.0% presented only occurrence and numeric frequencies, and 25.0% had no quantitative methods. The majority of the studies (62.5%) was conducted in southeastern Brazil. At least 89 species were reported as *S. guianensis*' preys: 78 species of fishes, 7 of cephalopods and 4 of crustaceans. From all evaluated studies,