Vol. 18: 63–68, 2013
doi: 10.3354/ab00491

AQUATIC BIOLOGY
Aquat Biol

Published online March 5

Reference ranges for body condition in wild bottlenose dolphins *Tursiops truncatus*

Leslie B. Hart[1,*], Randall S. Wells[2], Lori H. Schwacke[1]

[1]National Oceanic and Atmospheric Administration, National Centers for Coastal Ocean Science, Hollings Marine Laboratory, 331 Fort Johnson Road, Charleston, South Carolina 29412, USA

[2]Chicago Zoological Society, c/o: Mote Marine Laboratory, 1600 Ken Thompson Parkway, Sarasota, Florida 34236, USA

ABSTRACT: Marine mammal body condition, as evaluated by a combination of mass, length, and/or girth measurements, is considered an indicator of nutritional status. We used measurements of total mass, total length, and maximum girth from long-term bottlenose dolphin *Tursiops truncatus* capture–release research conducted in Sarasota Bay, Florida, USA, (1987 to 2009) to develop 95th percentile reference ranges for 2 body condition models: (1) total mass versus total length and (2) maximum girth versus total length. Nonlinear and linear quantile regression methods were used to estimate the parameters for the reference ranges and develop predictive models to examine body condition among individual dolphins. The flexibility of these models and reliance upon commonly acquired morphometrics allows for broad application among researchers lacking data on mass or age. Ultimately, these reference ranges can be used to evaluate and compare the body condition of individual animals and provide an additional metric for evaluating the general health of coastal populations.

KEY WORDS: Marine mammal · Morphometrics · Health assessment · Nutritional condition · Mass · Girth · Length · Quantile regression · Sarasota Bay

INTRODUCTION

Body size and growth in marine mammals have been assessed using a variety of morphometrics, including direct measurements of length, girth, and mass (Lockyer & Morris 1987, Read et al. 1993, Hammill et al. 1995, Trites & Pauly 1998, Mueller et al. 2011), body volume and mass based on length and axillary girth (Innes et al. 1981, Castellini & Calkins 1993), weight-to-length ratio (Ridgway & Fenner 1982, Mueller et al. 2011), blubber mass (Read 1990), blubber mass and skin thickness relative to body length and girth (Pitcher et al. 2000), and anatomical measurements from aerial photographs (Perryman & Lynn 2002, Miller et al. 2012). Direct measurements of length, mass, and girth contribute to the determination of body size; however, body size should not be confused with estimates of body condition or shape. Body condition, which is often evaluated using combinations of mass, length, blubber, and girth measurements, can be used as an indicator of the nutritive condition of individuals and populations (Ridgway & Fenner 1982, Read 1990, Harwood et al. 2000, Perryman & Lynn 2002, Mueller et al. 2011, Miller et al. 2012). In humans, a body mass index (BMI) relating weight to height^2 is often used to identify underweight and obese people (Keys et al. 1972, NIH 1998). Similarly, in harp and ringed seals, a BMI based on empirical data and a ratio of weight to an exponentiation of length, where the exponent is a fitted parameter, has been used to examine overall body condition and temporal changes that may reflect of differences in nutritional condition (Hammill et al. 1995, Harwood et al. 2000).

Many studies of body size and condition have employed ordinary least squares (OLS) regression methods to examine relationships among different metrics (Read 1990, Castellini & Calkins 1993, Trites & Pauly 1998, Pitcher et al. 2000, Mueller et al. 2011);

*Email: leslie.burdett@noaa.gov

however, mean regression estimates do not necessarily reflect the variation surrounding individual observations (Koenker & Hallock 2001, Cade & Noon 2003). Quantile regression methods (Koenker & Bassett 1978) provide a way to model and estimate the responses for the entire distribution of the dependent variable, and allow us to predict responses within a specified interval of the distribution (e.g. 95th percentile; Koenker & Hallock 2001, Cade & Noon 2003).

The objective of our study was to use long-term morphometric data and quantile regression to examine relationships between measurements that indicate an individual's body condition (i.e. mass, length, maximum girth), and develop corresponding 95th percentile reference ranges for wild bottlenose dolphins. Reference ranges were developed for 2 different body condition models to accommodate differences in morphometric data availability. Both models are age-independent, which allows for broader applicability, as age determination for bottlenose dolphins requires physical handling, invasive procedures, and can be costly and time consuming. Baseline reference ranges provide a foundation for comparing the health status of individuals and stocks to a reference population (e.g. hematological and serum biochemical variables; Schwacke et al. 2009). Because body condition reflects nutritional status, it is anticipated that these reference ranges can identify individuals in poor nutritive condition during population health assessments.

MATERIALS AND METHODS

Bottlenose dolphins in Sarasota Bay, Florida (USA), have been studied since 1970, and are considered to be part of a demographically closed, long-term resident population of approximately 160 individuals (Wells 2009). Health parameters, morphometrics, and ages are known for many individuals in the population through capture–release efforts (Wells et al. 2004). For this study, we used sex, pregnancy status (determined by ultrasonography), and measurements of total length (TL), maximum girth (MG), and total mass (TM) from capture–release projects conducted in May to July 1987 to 2009. TL was measured to the nearest mm from upper rostrum tip to fluke notch (Read et al. 1993), MG (to the nearest mm) was measured as the circumference of the body just anterior to the dorsal fin (Tolley et al. 1995), and TM was determined to the nearest 0.1 kg using a scale or load cell onboard a research vessel (Read et al. 1993).

We used the software packages R 2.15.0 (R Foundation for Statistical Computing; http://cran.r-project.org/) and Statistica 9.0 (StatSoft; http://www.statsoft.com/) for statistical analyses. Adult male and female bottlenose dolphins in Sarasota Bay are sexually dimorphic in TL, MG, and TM (Read et al. 1993, Tolley et al. 1995); therefore, analyses were conducted separately for each sex. Pregnant females were excluded from all analyses due to changes in body size that result from carrying a fetus. Analyses were restricted to summer observations due to potential seasonal differences in body condition resulting from changes in blubber depth in response to warmer water temperatures (Meagher et al. 2008). Reference ranges were not developed for other seasons due to limited sample sizes. To avoid repeated observations of individuals, only the most recent measurements for an individual were included for regression models. Selecting the most recent measurement for an individual also helped to build the sample size for larger animals, providing a more even distribution across length classes. Also, dolphins measured during 2005 and 2006 were excluded due to a red tide harmful algal bloom that significantly reduced prey availability and impacted the nutritional status of Sarasota Bay dolphins (Gannon et al. 2009, Powell & Wells 2011).

TL and TM were available for 88 male and 72 female dolphins, and MG measurements were available for 86 males and 70 females. Ranges and means for TM, MG, and TL were greater for males than females. Nonlinear OLS regression was used to examine the relationship between TM and TL as proposed by Innes et al. (1981) (Model 1):

$$TM = 10^a \times TL^b \tag{1}$$

where a and b are estimated parameters. Nonlinear quantile regression was subsequently used to develop 95% reference intervals for this model. From Eq. (1), a BMI can be calculated (Hammill et al. 1995, Harwood et al. 2000):

$$BMI = \frac{TM}{TL^b} \times 10000 \tag{2}$$

and can be used to compare across populations or between time periods (Harwood et al. 2000).

Measurements of TM are not always possible, as it requires the physical lifting of individuals and use of a load cell or scale. Therefore, in addition to the mass-dependent reference range, we constructed a reference interval that could be used as a proxy for body condition in the absence of data on mass. Although Miller et al. (2012) found significant fluctuations in body width at posterior portions of adult female right whales, other studies of girth among wild and captive cetaceans using direct measurements and aerial photographs indicate that measurements at the widest part of the body fluctuate more with seasons, changes in nutritional condition, and growth (Lockyer & Morris 1987, Perryman & Lynn 2002). For both males and females, TL and MG in the present study were significantly correlated (Fig. 1), so length-dependent MG reference ranges were developed using linear quantile regression methods and the following function (Model 2):

$$MG = a + b \times TL \qquad (3)$$

OLS regression methods were used to evaluate the fit of these models based on statistical significance of parameter estimates, distribution of the residuals versus fitted values, and R^2 values (Table 1). We developed 95% reference intervals using quantile regression methods for both body condition models. Non-linear modeling is an iterative process for parameter estimation; therefore, initial values for the median quantile regression model were based on parameter estimates produced by OLS models (Table 1). Similarly, median quantile parameter estimates were used as initial values for the upper and lower quantile modeling. For both sexes, parameter estimates for the upper (τ = 0.975) and lower (τ = 0.025) 95th quantiles (Table 2) were used to plot 95th percentile reference ranges for both body condition models (Fig. 2).

Model 1 can be used to evaluate the body condition of an individual animal by comparing the individual's measured mass (TM_{actual}) to predicted estimates of total mass (PTM) for the upper and lower 95th quantiles (Table 2). An animal would be designated as having poor body condition (i.e. below the lower 95th threshold) if:

$$TM_{actual} < PTM_{0.025} = 10^{a_{0.025}} \times TL_{actual}^{b_{0.025}} \qquad (4)$$

where TL_{actual} is the measured total length of the individual, and $PTM_{0.025}$ is the predicted total mass at the lower 95th percentile threshold based on the parameter estimates for *a* ($a_{0.025}$) and *b* ($b_{0.025}$) from the lower quantile model (Table 2). Similarly, Model 2 can be used to evaluate poor body condition based on maximum girth measurements:

$$MG_{actual} < PMG_{0.025} = a_{0.025} + b_{0.025} \times TL_{actual} \qquad (5)$$

where MG_{actual} is the measured girth of an individual, TL_{actual} is the individual's measured total length, and $PMG_{0.025}$ is the predicted maximum girth based on lower quantile parameter estimates ($a_{0.025}$, $b_{0.025}$) for Model 2 (Table 2).

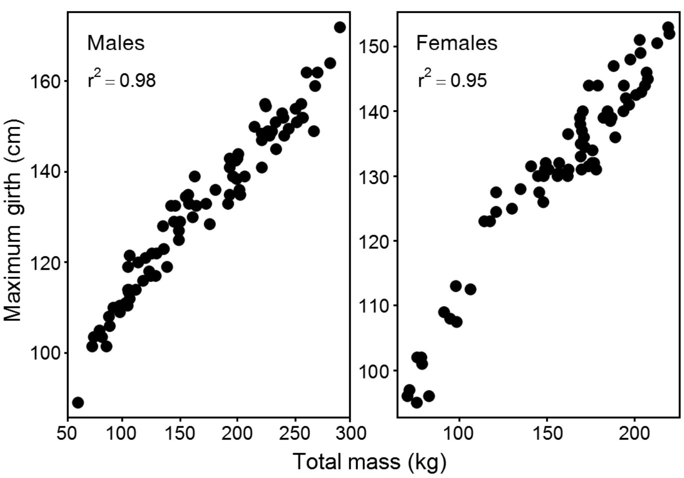

Fig. 1. *Tursiops truncatus*. Maximum girth vs. total mass for bottlenose dolphins captured and released in Sarasota Bay, Florida, USA (1987–2009)

Table 1. Ordinary least squares (OLS) parameter estimates (± SE) and R^2 values of body condition models for bottlenose dolphins. Model 1: $TM = 10^a \times TL^b$; Model 2: $MG = a + b \times TL$. TM: total mass, TL: total length, MG: maximum girth

	Model	*a*	*b*	R^2
Males	1	−5.40 ± 0.27	3.20 ± 0.11	0.93
	2	−14.71 ± 6.33	0.62 ± 0.03	0.86
Females	1	−4.29 ± 0.30	2.73 ± 0.12	0.91
	2	−5.68 ± 7.82	0.58 ± 0.03	0.82

Table 2. Parameter estimates (± SE) for median, and upper and lower 95th quantiles of body condition Model 1 ($TM = 10^a \times TL^b$) and Model 2 ($MG = a + b \times TL$) for bottlenose dolphins. TM: total mass, TL: total length, MG: maximum girth

	Quantile	τ	*a*	*b*
Model 1				
Males	Upper	0.975	−5.47 ± 0.62	3.27 ± 0.27
	Median	0.5	−5.60 ± 0.47	3.29 ± 0.20
	Lower	0.025	−5.52 ± 0.40	3.23 ± 0.17
Females	Upper	0.975	−4.57 ± 0.55	2.88 ± 0.23
	Median	0.5	−4.62 ± 0.49	2.87 ± 0.21
	Lower	0.025	−5.03 ± 0.28	3.01 ± 0.12
Model 2				
Males	Upper	0.975	−15.75 ± 19.59	0.70 ± 0.09
	Median	0.5	−21.45 ± 8.93	0.65 ± 0.04
	Lower	0.025	−12.06 ± 4.73	0.57 ± 0.02
Females	Upper	0.975	18.38 ± 23.56	0.53 ± 0.10
	Median	0.5	−7.47 ± 15.48	0.59 ± 0.06
	Lower	0.025	2.50 ± 8.22	0.50 ± 0.04

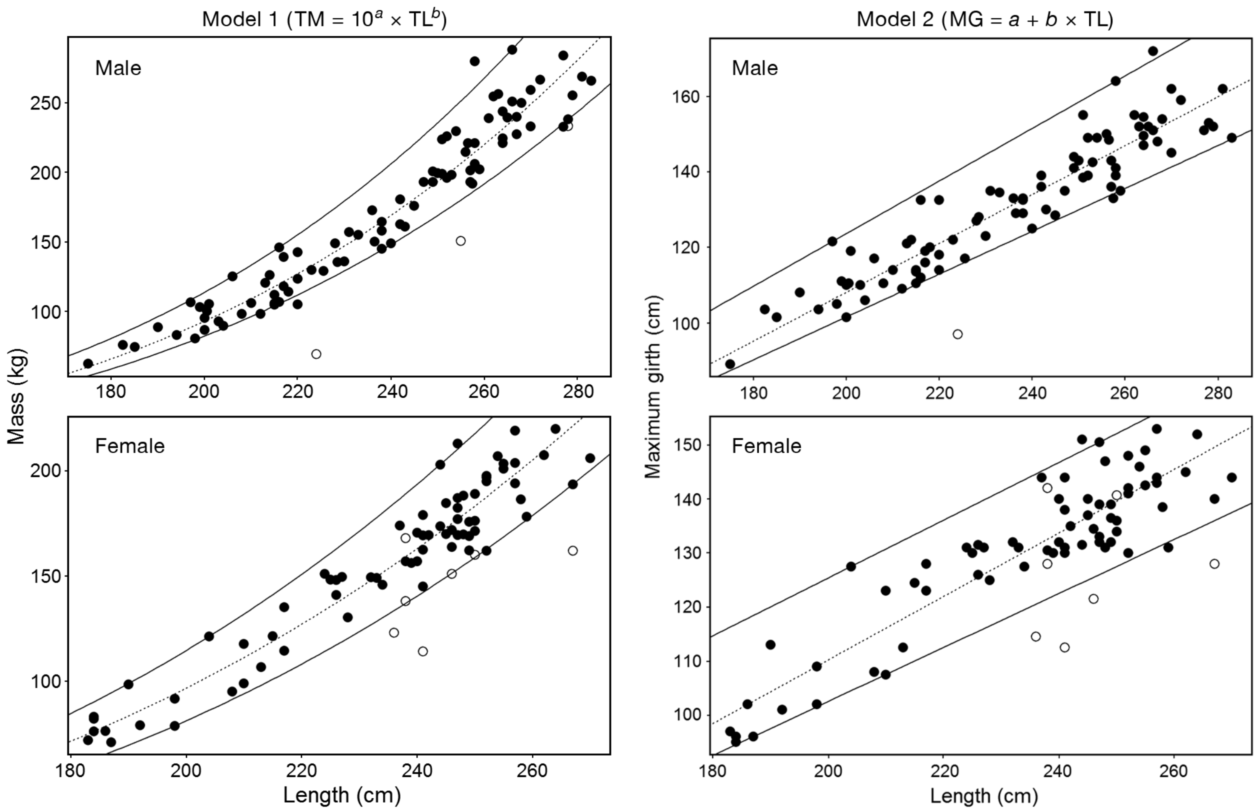

Fig. 2. *Tursiops truncatus*. 95% reference ranges for body condition for dolphins captured and released in Sarasota Bay, Florida. Solid lines: upper and lower 95th quantiles; dotted line: median. Closed circles: live dolphins; open circles: carcasses. TM: total mass, TL: total length, MG: maximum girth (MG)

RESULTS AND DISCUSSION

To demonstrate the applicability of the reference ranges presented here, we examined the TM, TL, and MG of recently stranded resident dolphins (11 males, 17 females). Although the body condition of a stranded dolphin would depend on the cause of stranding (e.g. chronic disease/debilitation versus acute trauma), we would expect a higher likelihood of poor body condition for stranded individuals. Of the 6 male carcasses with complete data for evaluating body condition, 3 were excluded because they

Table 3. *Tursiops truncatus*. Comparison of total mass (TM_{actual}), maximum girth (MG_{actual}), and body mass index (BMI) of stranded dolphins to 95th quantile thresholds for predicted total mass (PTM) and predicted maximum girth (PMG) based on 2 body condition models. **Bold**: TM and MG measurements are below the lower 95% thresholds. TL_{actual}: total length

Dolphin	TM_{actual} (kg)	TL_{actual} (cm)	MG_{actual} (cm)	BMI	Model 1		Model 2	
					$PTM_{0.025}$	$PTM_{0.975}$	$PMG_{0.025}$	$PMG_{0.975}$
Males								
1	69.0	224.0	97.0	0.018[a]	**117.8**	164.2	**115.6**	141.1
2	233.5	278.0	NA	0.030[a]	**236.7**	332.7	NA	NA
3	150.5	255.0	NA	0.025[a]	**179.1**	250.8	NA	NA
Females								
4	168.0	238.0	142.0	0.118	136.9	189.0	121.5	145.7
5	160.0	250.0	140.7	0.097	158.8	217.7	127.5	152.1
6	162.0	267.0	128.0	0.080[a]	**193.6**	263.1	**136.0**	161.2
7	138.0	238.0	128.0	0.097	136.9	189.0	121.5	145.7
8	114.0	241.0	112.5	0.077[a]	**142.2**	195.9	**123.0**	147.3
9	123.0	236.0	114.5	0.089[a]	**133.4**	184.4	**120.5**	144.6
10	151.0	246.0	121.5	0.096	**151.2**	207.8	**125.5**	150.0

[a]Estimated BMI is below the predicted lower quantile ($BMI_{0.025} = 10^{a_{0.025}} \times 10\,000$; males = 0.030, females = 0.093; Table 2)

were calves with lengths below the predictive range of the model (i.e. <175 cm). The remaining 3 dolphins had poor body condition (Fig. 2). TM and MG for Dolphin 1 were below the 95% reference interval for both body condition models (Eqs. 4 & 5; Table 3, Fig. 2). Similarly, the estimated BMI (Eq. 2) was below the lower quantile parameter estimate for Model 1 (Table 3). At necropsy, this animal was declared emaciated and had indications of infection and human-induced trauma. Dolphins 2 and 3 poor body condition by Model 1 (Eq. 4; Table 3, Fig. 2); 1 was emaciated at necropsy and both had evidence of human trauma including ingestion of fishing gear. These 2 animals were not evaluated using Model 2, because maximum girth measurements were not recorded. The body condition of 7 fresh-stranded female dolphins was evaluated using both models and the equation for BMI. Dolphins 6, 8, 9, and 10 had poor body condition by both models, and 3 had low BMI estimates (Table 3, Fig. 2). The cause of death was undetermined for 1 of these dolphins, while the remaining 3 had indications of stingray barb punctures to the lung, 2 were considered emaciated at necropsy, and 1 presented evidence of hook and line ingestion. Stingray barb injury to the lung was also suspected for 2 of the females near the lower threshold for Model 1 (Fig. 2). Stingray barb puncture and fishing gear ingestion do not always cause acute fatality; however, swallowed fishing gear can inhibit prey consumption, leading to weight loss and emaciation, while stingray barbs can cause organ damage and systemic infection (Walsh et al. 1988), thus negatively affecting body condition.

These reference ranges were developed for individuals sampled during the warmest months of the year, and caution should be used when applying them to stocks or populations sampled during winter months. However, a subset of male and female dolphins sampled in February 2003 to 2005 (9 males, 15 females) fell within or slightly above the reference ranges except for 1 female that was below the lower 95th threshold for Model 2 (not shown). The reference ranges developed here used data from estuarine dolphins inhabiting a bay on the western coast of Florida. Read et al. (1993) found size similarities between dolphins sampled in Sarasota Bay and the western North Atlantic, suggesting that these reference ranges are likely applicable to populations from other regions of the USA. As bottlenose dolphin health assessment projects in the USA have increased in number and geographic range in recent years (Hansen et al. 2004, Schwacke et al. 2010, 2012), these reference ranges can be used to identify individuals or stocks with compromised health conditions that may be related to or reflected by changes in body condition.

Acknowledgements. We thank B. Irvine, M. Scott, and the staff, interns, and volunteers of the Sarasota Dolphin Research Program (SDRP) for ensuring the safety and effectiveness of the capture–release projects. Sarasota Bay stranding data were provided by G. Lovewell of Mote Marine Laboratory. Thanks also to M. Neely, S. Lane, and T. Speakman for reviewing this manuscript. NOAA and Dolphin Quest provided much of the support for health assessments. Capture–release work was conducted under NOAA Scientific Research Permits and IACUC approvals issued to R.S.W. Funding for data analysis was provided by NOAA's Center of Excellence for Oceans and Human Health at the Hollings Marine Laboratory and the Office of Naval Research's Marine Mammals and Biology Program.

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Editorial responsibility: Brent Stewart, San Diego, California, USA

Submitted: June 27, 2012; Accepted: January 2, 2013
Proofs received from author(s): February 21, 2013