Stressed and slim or relaxed and chubby? A simultaneous assessment of gray whale body condition and hormone variability

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Of all mammals, baleen whale physiology is one of the most poorly understood due to many complications that arise during sampling (Hunt *et al.*, 2006, 2013). The size, brief surfacing behavior, and aquatic environment of whales are all challenging for sample collection. This knowledge gap in baleen whale physiology limits the ability to understand how baleen whale health varies due to intrinsic (e.g., age, reproductive stage) and extrinsic (e.g., prey limitation, disturbance) factors. However, a number of techniques using multiple matrices have recently been developed that enable nonlethal and minimally invasive (or noninvasive) physiological monitoring. For instance, analysis of cetacean feces, respiratory vapor (i.e., blow), and blubber biopsy samples can be used to monitor hormones and respiratory microbiome (Hunt *et al.*, 2013). Additionally, photogrammetric analysis of aerial drone images can be used to assess whale body condition as an indicator of nutritional state (Christiansen *et al.*, 2018; Burnett *et al.*, 2019; Soledade Lemos *et al.*, 2020). While all these techniques individually provide slices of information on baleen whale health, if these approaches are employed simultaneously, more robust and complete assessments of whale health may be feasible, including the potential to cross-validate methods.

Studies that employ simultaneous multidisciplinary techniques in marine mammal physiology are still scarce and have primarily assessed endocrine responses in association with contaminant loads (Haave *et al.*, 2003; Yordy *et al.*, 2010; Galligan *et al.*, 2019). Only a few studies have assessed hormone variation in relation to body condition. A study of Steller sea lion (*Eumetopias jubatus*) physiology found that mass loss during periods of energy restriction was negatively correlated with cortisol and positively correlated with insulin-like growth factor-I (IGF-I; Jeanniard du Dot *et al.*, 2009). When the sea lions were fed, cortisol decreased and IGF-I increased, suggesting that these two hormones are inversely correlated, together providing good indicator of nutritional stress in sea lions. Thyroid hormones (i.e., triiodothyronine - T3 and thyroxine - T4) were also simultaneously monitored during this study yet displayed no correlations with changes in body condition (Jeanniard du Dot *et al.*, 2009). Another study examined correlations between Weddell seal (*Leptonychotes weddellii*) body condition and hormones that influence energy allocation, including cortisol, growth hormone, IGF-I and thyroid hormones (Shero *et al.*, 2015). Triiodothyronine was negatively correlated with body mass in reproductive females while T4 exhibited a positive relationship with body mass in nonreproductive females. To date, no study has evaluated associations between hormone variability and body condition in baleen whales, which limits the ability to understand physiological response to periods of prey limitation. As our oceans continue to change due to increased anthropogenic activities and climate change, a better understanding of connections between body condition and hormones will assist efforts to mitigate impacts at an individual and population level.

In this study, we employed simultaneous multidisciplinary techniques to investigate the effects of body condition variability on hormones associated with stress (i.e., cortisol) and energy allocation (i.e., cortisol and thyroid hormones) in Eastern North Pacific (ENP) gray whales (*Eschrichtius robustus*). The ENP population annually migrates from its southern breeding grounds in Baja California, Mexico, to its northern feeding grounds in the Bering and Chukchi seas (Calambokidis *et al.*, 2002). However, a group of ~200-250 individual ENP gray whales, known as the Pacific Coast Feeding Group (PCFG), does not perform the full migration and can be found along the coasts from northern California to southeastern Alaska during the summer (Calambokidis *et al.*, 2002; Calambokidis and Perez, 2017). These whales are frequently located within 10 km of the shore (Mate and Urban-Ramirez, 2003) and thus, are subject to numerous anthropogenic pressures, including coastal pollution, ambient noise, fishing gear entanglement, and vessel/ship strikes (Baird *et al.*, 2002; Moore and Clarke, 2002; Jones *et al.*, 2012). At the time of writing, the ENP population is under an unusual mortality event that resulted in 430 strandings of dead gray whales as of April 5, 2021 (2019-2021; NOAA, 2021). Therefore, it is of critical importance to assess the physiological state of this population simultaneously with environmental data in order to identify potential causes for unusual events such as this. In fact, a previous study on this same population hypothesized a link between poor whale body condition and low prey availability in the three years preceding the event (Soledade Lemos *et al.*, 2020).

Cortisol plays a role in lipid metabolism associated with unpredictable life situations, including energy intake shortages (Kitaysky *et al.*, 2001; Reeder and Kramer, 2005). Additionally, thyroid hormones are known to be influenced by caloric intake in terrestrial mammals, typically with decreases in the hormone concentrations when food is deprived (Nelson, 2005; Behringer *et al.*, 2018) - though these relationships might differ in marine mammals (Atkinson *et al.*, 2015). We hypothesize that cortisol and thyroid response would vary with whale body condition and therefore reflect energetic intake. An increase in cortisol and decrease in thyroid hormones were expected with poor body condition, and therefore a negative relationship between cortisol and thyroid hormones was also hypothesized.

We used a small research vessel (5.4 m rigid-hulled inflatable boat) to locate PCFG gray whales in their feeding ground along the Oregon coast, USA, between June and October of 2016, 2017, and 2018. All sampled whales were located within 10 km from the shore and photographs of left and right-hand sides and flukes were taken of each whale for photo-identification analysis. The number of sighted whales and calf presence was also recorded. If weather conditions were appropriate (e.g., good visibility and absence of strong winds) and whales were not showing any change in behavior due to the presence of the research vessel, drone-based videos were recorded for photogrammetry analysis (Burnett *et al.*, 2019; Soledade Lemos *et al.*, 2020). In order to correct barometric altimeter errors during data postprocessing, a calibration object of 1.0 m in length was centered in the frame of the drone camera during the beginning of all flights and recorded from 10 to 40 m of altitude. Drones used in this study included a DJI Phantom 3 Pro in 2016, a Phantom 4 in 2017, and a Phantom 4 Pro in 2018. All videos were recorded at a minimum altitude of 25 m and no behavioral responses of whales to the drone were noted.

In addition, fecal samples were opportunistically collected during these whale sightings using two 300 µm nylon mesh dipnets (methods described in Lemos *et al.*, 2020). Samples were transferred to sterile plastic jars and put on ice until stored in a freezer (-20°C) for later analysis. All samples were freshly voided from individuals that were also photographed, thus the samples could be attributed to a specific individual. Information on location, date, and time, were also recorded for all collected fecal samples.

Photo-identification analysis was conducted in Adobe Bridge (version 8.0.1.282), and photographs were compared to long-term gray whale photo-identification catalogs held by the Marine Mammal Institute at Oregon State University and Cascadia Research Collective (Olympia, WA). This comparison enabled the matching of lateral and fluke body marks and pigmentations of the same individual whale, and the retrieval of individual whale sighting histories that included information on sex and minimum age. If a whale was not found in the catalogs, the sex was determined through fecal genetic analyses (full methods described in Soledade Lemos *et al.*, 2020).

The photogrammetric analysis uses a three-program analysis suite to produce 11 morphometric attributes from drone imagery of the whale, while compensating for lens distortion and correcting for scaling errors (Burnett *et al.*, 2019; Soledade Lemos *et al.*, 2020). In short, images of whales lying straight and flat at the surface were extracted from video recordings and categorized as either good or poor image quality based on attributes relative to the whale body position, camera focus, and environmental conditions. If the image was scored as good, a series of whale body length and width measurements were conducted. From these measurements, we produced an estimate of the Body Area Index (BAI) value, which describes the relative body condition for each whale. Body Area Index is conceptually similar to the Body Mass Index (BMI) used in humans and is a unitless and length-invariant metric of body condition, estimated by normalizing two-dimensional body surface area by length (Burnett *et al.*, 2019). Thus, BAI enables comparisons of body condition among individual whales of different demographic units and lengths (i.e., calves:adults or males:females). A threshold of 5% coefficient of variance (CV) was applied for both whale length and BAI measurements to improve accuracy. Based on fieldwork observations, photo-identification, and photogrammetry results, each identified whale was assigned to a demographic unit based on sex and maturity in that specific year (e.g., pregnant in one year and resting in the following year; for demographic grouping details see Soledade Lemos *et al.*, 2020). To minimize the influence of demographic unit on correlations between body condition and hormones, we only included mature males and females (i.e., nonpregnant and nonlactating; identified through field observations/sighting history) in the following data analysis. These demographic units also had the largest sample sizes.

Fecal samples containing saltwater were filtered using unbleached coffee filters (Lemos *et al.*, 2020). Deionized water was then added to the samples, which were centrifuged for 10 minutes at 3,000 rpm (i.e., 1,000 RCF – relative force [g]) so as to extract salt remaining in the samples. The overlaying water was removed by pipetting and samples were frozen until lyophilized for 72 hr to remove all water content. All samples were analyzed for hormone metabolite concentration within 11 months of collection. Samples were weighed to the nearest 0.001 g (mean: 0.12 g, standard deviation: 0.07 g) and samples below 0.02 g were excluded from the analysis in order to avoid inflated values (Ayres *et al.*, 2012; Lemos *et al.*, 2020). When multiple samples from the same individual were collected on the same day, samples with higher mass were used in the analysis (Ayres *et al.*, 2012). Fecal hormone metabolites (HM; i.e., hormones metabolized in the gut before excretion in feces) were extracted following the procedure described in Lemos *et al.*, (2020). Briefly, extraction occurred by adding 90% methanol to the samples using binned solvent:sample ratios within a range of 1:10 to 1:25. Tubes were loaded onto a plate shaker at 500 rpm for 30 min, followed by centrifugation at 2,200 rpm for 20 min (i.e., 285 RCF), recovery of supernatant, and dilution for assay. Commercial Enzyme-linked Immunosorbent Assay (ELISA) kits for cortisol (Enzo Life Sciences, catalog #ADI-900-071, https://www.enzolifesciences.com) and T3 (Arbor Assays, catalog #K056-H1, https://www.arborassays.com) were used to assess glucocorticoid metabolite (GCm) and thyroid metabolite (Tm) concentrations, respectively. GCm were quantified in the three years of study, while Tm was only quantified in 2017 and 2018. Samples were run in duplicates in 2016 and 2017, and in triplicates in 2018. In order to maximize accuracy in our study, we repeated HM analysis in any samples with CV higher than 15% and/or percent bound outside the 15% and 85% range, until appropriate values were reached. Values below the limit of detection (<LOD; Cortisol: 0.156 ng.g^{-1}, T3: 0.078 ng.g^{-1}) were excluded from the analysis (Wood *et al.*, 2011). Information on cross-reactivities, assay sensitivity, intra- and interassay coefficient of variance percentage, as well as analytical and physiological validations of gray whale fecal samples were previously conducted and described in (Lemos *et al.*, 2020).

To correlate body condition and hormone variation in individual whales, BAI measurements and fecal HM data collected from the same whale on the same day were assessed in this study. However, there were times (n = 37) when drone flights were not conducted over whales on the same day a fecal sample was collected (i.e., due to weather or technical difficulties) or extracted images from the drone recordings did not pass our image quality control. Hence, to increase our sample size for correlation analysis, we used BAI values measured within ± 14 days of a fecal sample collection from the same individual as gray whales do not significantly change their body condition within a period of two weeks (paired *t*-test using all BAI values of individuals assessed within 14 days in 2016, 2017 and 2018: n = 61, p = 0.86, df= 60, *t* = - 0.174).

All statistical tests were conducted in R (version 3.5.0; R Core Team, 2019) and alpha < 0.05 were considered significant in all tests. Fecal hormone metabolites concentrations were log-transformed (log [value +1]) before statistical analysis. A residual analysis between body condition and HMs was initially conducted and any values greater than ± 2 standard deviations were considered outliers and excluded from subsequent analyses (n = 2). We conducted linear mixed models (LMM), using the *lme4* package in R (Bates *et al.*, 2015) to assess the effects of BAI, sex, year, minimum age, and day of the year (DOY) on both GCm and Tm concentrations. Glucocorticoid metabolites were also considered in Tm models, and vice versa. All models included the whale identity as random effect to account for pseudoreplication. In addition, DOY was either considered a fixed factor, so as to account for its direct influence on the models, or as a random factor, so as to account for variations in sampling per day. Model selection was based on the lowest Akaike's information criterion (AIC; Burnham *et al.*, 2011), and the fit of the selected models was evaluated using the marginal R^2 (R^2m; variance explained by fixed effects) and the conditional R^2 (R^2c; variance explained by both fixed and random effects) using the *MuMIn* package in R (Nakagawa and Schielzeth, 2013; Barton, 2020). We also used the *lmerTest* package (Kuznetsova *et al.*, 2017) to obtain *F*-statistics and *p*-values. Additional linear regressions between significant factors and response variables were conducted to verify the direction of the associations.

A total of 73 fecal samples from 17 different mature females (42 samples) and 19 different mature males (31 samples) were collected (Table 1; Fig. 1). However, 17 of these samples had no associated BAI information. In addition, 20 GCm and 24 Tm values were below LOD. These NA values for BAI and HMs were removed from the relevant statistical analysis, which reduced sample size to 32 (11 mature males and 10 mature females).

Twelve different models were run for each of the HM (Table 2). The model that best explained GCm variations was model 7, which included BAI, sex, year, and Tm (AIC = 63.71, df = 7, $R^2m = 0.43$, $R^2c = 0.43$). Within this model, BAI (F = 10.58, df = 27, $p < 0.01$; negative association) and year 2018 (F = 9.69, df = 27, $p < 0.01$) displayed significant effects on GCm. The model that best explained Tm variation was also model 7, which included BAI, sex, year, and GCm (AIC = 97.97, df = 7, $R^2m = 0.52$, $R^2c = 0.83$). Within this model, Tm was significantly affected by year 2018 ($F = 28.08$, df = 26, $p < 0.001$) and GCm (F = 5.45, df = 14, $p < 0.05$; positive association), where year was the most significant factor (Fig. 3). The R^2m values in both selected models indicate that 43% and 84% of the variance in GCm and Tm concentrations, respectively, is explained by the models; thus, other, unmeasured, factors also contribute to the variation of these hormones in gray whales, especially GCm. The R^2c and R^2m values for the GCm model were the same ($R^2c = 0.433$, $R^2m = 0.433$; Table 2), indicating that the random factor (i.e., whale identity) did not actually contribute to the GCm model. Conversely, the random factor of whale identity highly contributed to the Tm model ($R^2c = 0.522$, $R^2m = 0.835$; Table 2).

Linear regression results with all data grouped together (i.e., regardless of year, sex, age, or DOY) indicate a significant negative association between GCm and BAI (rate of change = -

0.15, $F_{1,37} = 17.55$, $R^2 = 0.303$, $p < 0.001$; Fig. 2a). No significant correlations were observed between Tm and BAI (rate of change = -0.21, $F_{1,33} = 2.96$, $R^2 = 0.054$, $p = 0.09$; Fig. 2b), or between Tm and GCm (rate of change = 0.37, $F_{1,42} = 1.10$, $R^2 = 0.002$, $p = 0.30$; Fig. 2c).

Our results indicate that gray whale body condition (BAI) was negatively correlated with GCm and displayed no significant correlation with Tm. These findings suggest that GCm play an important role in energy allocation in gray whales, while Tm may not be as directly associated (though note that statistical power was limited in this study). Even though Tm did not display a significant correlation with BAI, *p*-value was relatively low (<0.10) and the yearly trends were negative, which is also explained by the positive correlation between Tm and GCm. Results suggest a relationship may exist between Tm and BAI that could be resolved with greater sample size (i.e., greater statistical power) in future studies; thus, we recommend further research on Tm and body condition.

A similar pattern between body condition and hormones was previously reported in Steller sea lions, in which increased blood cortisol levels were detected during periods of energy restriction and body mass reduction, and no significant correlations with thyroid hormones were observed (Jeanniard du Dot *et al.*, 2009). In contrast, other studies documented a negative correlation between thyroid hormones and body mass in Weddell seals (Shero *et al.*, 2015), and between Tm and food intake in killer whales (*Orcinus orca*; Ayres *et al.*, 2012).

The year of 2018 was an important explanatory factor in both gray whale GCm and Tm models. According to Soledade Lemos et al. (2020), gray whales exhibited poor body condition in 2018, which was associated with two prior years of poor local upwelling conditions that may have caused reduced prey availability. Therefore, gray whales in 2018 may have endured prolonged nutritional stress leading to the higher GCm concentrations detected in that year. The significance of 2018 in the variation of Tm is less clear given the lack of significant correlation between Tm and BAI. Therefore, it is likely that factors other than, or in addition to, body condition affect Tm concentrations in gray whales. Thyroid hormones are involved in multiple metabolic activities, including thermoregulation and carbohydrate utilization (Behringer *et al.*, 2018). In some mammals, thyroid hormone variation can be driven by thermoregulatory needs, with thyroid hormones rising if animals are chilled and/or thin (poorly insulated; Oki and Atkinson, 2004). However, cetaceans may not experience strong variation in thermoregulatory energetics due to their effective insulation (Hokkanen, 1990). In fact, it has been suggested that some cetaceans are able to conserve body heat in subfreezing waters and thus migrations may not be necessary due to thermoregulatory requirements (e.g., Sumich, 1986; Pitman *et al.*, 2019). Instead, migration may occur due to calf-predator encounter avoidance (Payne, 1995; Corkeron and Connor, 1999; "calf refuge hypothesis"; Connor, 2001). Alternatively, the increase in gray whale thyroid hormones may be associated with accelerated carbohydrate utilization, which appears to be associated with increased demand for adenosine triphosphate (ATP), the content of carbohydrate in the diet, and the nutritional state of the animal (Goodman, 2009). Thus, it is possible that the poor nutritional state and/or the deficit in carbohydrates in gray whales' diet in 2018 caused whales to produce higher Tm levels to metabolize stored carbohydrates as an energy source.

It is important to highlight that we only quantified Tm in two years, thus our sample size was relatively small (n = 49) and potentially limited the ability to determine drivers of Tm variability. Additionally, knowledge on how thyroid hormones behave in baleen whales is restricted to findings from just a few studies, including blow samples of North Atlantic right whales (*Eubalaena glacialis*; Hunt *et al.*, 2014), baleen samples of multiple species (Hunt *et al.*, 2017; Lysiak *et al.*, 2018), and fecal samples of humpback whales (*Megaptera novaeangliae*; Hunt *et* *al.*, 2019) and gray whales (Lemos *et al.*, 2020). Therefore, we recommend further monitoring on both Tm and GCm concentrations in relation to baleen whale body condition, prey availability, respiratory rates, behavior, and other such factors known to influence Tm levels, such as sex, age, reproductive state, season, and migration (Hunt *et al.*, 2019).

In this study we demonstrate the added value and knowledge gained through simultaneous collection of physiological data from baleen whales over time. Although a larger sample size would improve the power of our analyses and results, we were still able to identify links between body condition and hormones associated with stress and energy allocation, including a negative correlation between GCm and BAI, and a positive correlation between GCm and Tm. The former relationship demonstrates that gray whales can indeed be "stressed" and "slim" or "relaxed" and "chubby". The latter relationship demonstrates that thyroid hormones may not be highly involved with energy intake, counter to our initial hypothesis. Rather, Tm may be related to other common metabolic activities such as carbohydrate utilization.

These findings indicate that the use of simultaneous gray whale physiology data is a useful tool for a better understanding of whale bioenergetic strategies to cope with predictable and unpredictable dietary shifts. Such information is paramount when developing conservation policies and is, therefore, crucial in the protection of the species.

Continued monitoring of body condition and hormone levels of this gray whale population will generate 'health profiles' of individual whales, enabling assessment of change over time and potential identification and diagnosis for variations in population health. Furthermore, these demographically informed health profiles would contribute to an improved understanding of baleen whale physiology, allowing inference for other baleen whale populations, where collection of these physiological data is more challenging.

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Tables

Table 1: Summary of body condition and fecal hormone results by demographic unit compared in this study. Number of observations (N_{obs}), number of individuals (N_{ind}), mean concentration ± standard deviation (sd), median and range (min – max) of glucocorticoid metabolites (GCm; $ng.g^{-1}$, dried mass, thyroid metabolites (Tm; $ng.g^{-1}$, dried mass) and Body Area Index (BAI) by demographic unit.

	N_{obs}	N_{ind}	**GCm mean ± sd** **median** **range**	**Tm mean ± sd** **median** **range**	**BAI mean ± sd** **median** **range**
Mature males	31	19	19.10 ± 10.89 19.76 3.81 – 41.86	195.01 ± 472.94 43.28 1.28 – 2,134.86	40.31 ± 3.18 39.87 35.54 - 47.00
Mature females	42	17	15.20 ± 9.09 13.83 4.77 – 48.30	109.68 ± 152.97 63.55 3.99 – 624.50	39.91 ± 1.83 39.82 36.56 – 43.62

Table 2: Linear mixed model selection parameters of gray whale glucocorticoid metabolite (GCm) and thyroid metabolite (Tm) concentrations relative to Body Area Index (BAI), sex, year, and day of the year (DOY). GCm is also considered in the Tm model and vice versa. All models used whale identification (ID) as a random effect. Models alternatively use DOY as random or fixed effects to verify its relative importance to the models as a fixed factor or to account for variations in sampling per day as a random factor. Model in bold represents the chosen model based on the lowest Akaike Information Criterion (AIC). The number of observations for each model is indicated by N_{obs} and the degrees of freedom by DF. The fit of the selected models is represented by the marginal R^2 (R^2m; variance explained by fixed effects) and the conditional R^2 (R^2c; variance explained by both fixed and random effects).

Models	N_{obs}	**DF**	**AIC**	R^2m	R^2c
Glucocorticoid metabolites (GCm) as the response variable:					
1) GCm ~ BAI + sex + year + (1|whale ID)	39	7	70.73191		
2) GCm ~ BAI + sex + year + DOY + (1|whale ID)	39	8	82.16471		
3) GCm ~ BAI + sex + year + (1|whale ID) + (1|DOY)	39	8	72.51293		
4) GCm ~ BAI + sex + year + age + (1|whale ID)	36	8	73.93764		
5) GCm ~ BAI + sex + year + age + DOY + (1|whale ID)	36	9	83.18066		
6) GCm ~ BAI + sex + year + age + (1|whale ID) + (1|DOY)	36	9	75.93764		
7) GCm ~ BAI + sex + year + Tm + (1|whale ID)	**32**	**7**	**63.71276**	**0.433**	**0.433**
8) GCm ~ BAI + sex + year + Tm + DOY + (1|whale ID)	32	8	74.13161		
9) GCm ~ BAI + sex + year + Tm + (1|whale ID) + (1|DOY)	32	8	65.71276		
10) GCm ~ BAI + sex + year + age + Tm + (1|whale ID)	29	8	66.84847		
11) GCm ~ BAI + sex + year + age + Tm + DOY + (1|whale ID)	29	9	74.52206		
12) GCm ~ BAI + sex + year + age + Tm + (1|whale ID) + (1|DOY)	29	9	68.84847		
Thyroid metabolites (Tm) as the response variable:					
1) Tm ~ BAI + sex + year + (1|whale ID)	35	6	113.94064		
2) Tm ~ BAI + sex + year + DOY + (1|whale ID)	35	7	124.04176		
3) Tm ~ BAI + sex + year + (1|whale ID) + (1|DOY)	35	7	115.78341		
4) Tm ~ BAI + sex + year + age + (1|whale ID)	32	7	109.76183		
5) Tm ~ BAI + sex + year + age + DOY + (1|whale ID)	32	8	120.21030		
6) Tm ~ BAI + sex + year + age + (1|whale ID) + (1|DOY)	32	8	111.76183		
7) Tm ~ BAI + sex + year + GCm + (1|whale ID)	**32**	**7**	**97.97389**	**0.522**	**0.835**
8) Tm ~ BAI + sex + year + GCm + DOY + (1|whale ID)	32	8	107.63614		
9) Tm ~ BAI + sex + year + GCm + (1|whale ID) + (1|DOY)	32	8	99.90223		
10) Tm ~ BAI + sex + year + age + GCm + (1|whale ID)	32	7	109.76183		
11) Tm ~ BAI + sex + year + age + GCm + DOY + (1|whale ID)	32	8	120.21030		
12) Tm ~ BAI + sex + year + age + GCm + (1|whale ID) + (1|DOY)	32	8	111.76183		

Figure captions

Figure 1: Frequency histograms of log (a) fecal glucocorticoid metabolite (GCm) and (b) thyroid metabolite (Tm) concentrations (ng.g^{-1}, dried mass), and (c) Body Area Index (BAI) by demographic units in gray whales sampled during May to October of 2016-2018 off the Oregon coast, USA. Individual whales may be represented multiple times in these plots as some were re-sighted within and between years.

Figure 2: Linear correlations between (a) Body Area Index (BAI) and glucocorticoid metabolites (GCm; ng.g^{-1}, dried mass), (b) BAI and thyroid metabolites (Tm), and (c) GCm and Tm by years in gray whales sampled during May to October of 2016-2018 along the Oregon coast, USA. Individual whales may be represented multiple times in these plots as some were re-sighted within and between years. Asterisks indicate significant correlations between the variables.

Figure 3: Boxplots of fecal glucocorticoid metabolite (GCm) and thyroid metabolite (Tm) concentrations (ng.g^{-1}, dried mass) by years of gray whales (mature females and males group together) sampled during May to October of 2016-2018 along the Oregon coast, USA. Individual whales may be represented multiple times in these plots as some were re-sighted within and between years. Asterisks indicate significant correlations between the variables. The line within the box is the median, the box encloses 25–75% of the data, the whiskers outside the box enclose 5–95% of the data, and filled circles are outliers.