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Original investigation

Seasonal response of ghrelin, growth hormone, and insulin-like growth factor I in the free-ranging Florida manatee (*Trichechus manatus latirostris*)

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ABSTRACT

Seasonal changes in light, temperature, and food availability stimulate a physiological response in an animal. Seasonal adaptations are well studied in Arctic, Sub-Arctic, and hibernating mammals; however, limited studies have been conducted in sub-tropical species. The Florida manatee (*Trichechus manatus latirostris*), a sub-tropical marine mammal, forages less during colder temperatures and may rely on adipose stores for maintenance energy requirements. Metabolic hormones, growth hormone (GH), insulin-like growth factor (IGF)-I, and ghrelin influence growth rate, accretion of lean and adipose tissue. They have been shown to regulate seasonal changes in body composition. The objective of this research was to investigate manatee metabolic hormones in two seasons to determine if manatees exhibit seasonality and if these hormones are associated with seasonal changes in body composition. In addition, age related differences in these metabolic hormones were assessed in multiple age classes. Concentrations of GH, IGF-I, and ghrelin were quantified in adult manatee serum using heterologous radioimmunoassays. Samples were compared between short (winter) and long (summer) photoperiods ($n = 22$ male, 20 female) and by age class (adult, juvenile, and calf) in long photoperiods ($n = 37$). Short photoperiods tended to have reduced GH ($p = 0.08$), greater IGF-I ($p = 0.01$), and greater blubber depth ($p = 0.03$) compared with long photoperiods. No differences were observed in ghrelin ($p = 0.66$). Surprisingly, no age related differences were observed in IGF-I or ghrelin concentrations ($p > 0.05$). However, serum concentrations of GH tended ($p = 0.07$) to be greater in calves and juveniles compared with adults. Increased IGF-I, greater blubber thickness, and reduced GH during short photoperiod suggest a prioritization for adipose deposition. Whereas, increased GH, reduced blubber thickness, and decreased IGF-I in long photoperiod suggest prioritization of lean tissue accretion. Hormone profiles in conjunction with difference in body composition between photoperiods indicate seasonal adjustments in manatee nutrient partitioning priorities.

Introduction

Mammals live in diverse environments and are confronted with consistent seasonal environmental fluctuations in light, temperature, and food availability. Seasonal environmental changes may stimulate a physiological response in an animal often initiated by the endocrine system to maintain homeostasis (Florant and Healy, 2012). Environmental stimuli may elicit a response of the organism to adjust homeostatic set points. These adjustments result in acclimatization and may include responses, such as alterations in food intake, body weight, fat content, and hormonal functions (Florant and Healy, 2012). Metabolic hormones such as ghrelin, growth hormone (GH), and insulin-like growth factor (IGF)-I function to regulate tissue-specific energy allocation and together link growth physiology, developmental age, energy balance (the amount of calories consumed versus metabolized and stored), and nutritional status (negative [fasting] or positive nutrient intake) in domestic animals (Bergan et al., 2015; Breier, 1999; Cammisotto et al., 2010; Castaneda et al., 2010; Lawrence and Fowler, 1997).

The secretion of the metabolic hormone ghrelin in the stomach is influenced by nutritional status (Cummings et al., 2001; Salbe et al., 2004). The increase of ghrelin during fasting or reduced nutrient intake stimulates the release of GH, fat oxidation, and food intake, and inhibits body weight gain and adiposity (Tschop et al., 2000). Growth hormone regulates energy deposition and mobilization through its direct action on adipose tissue (Bergan et al., 2015;

Abbreviations: GH, growth hormone; IGF, insulin-like growth factor.

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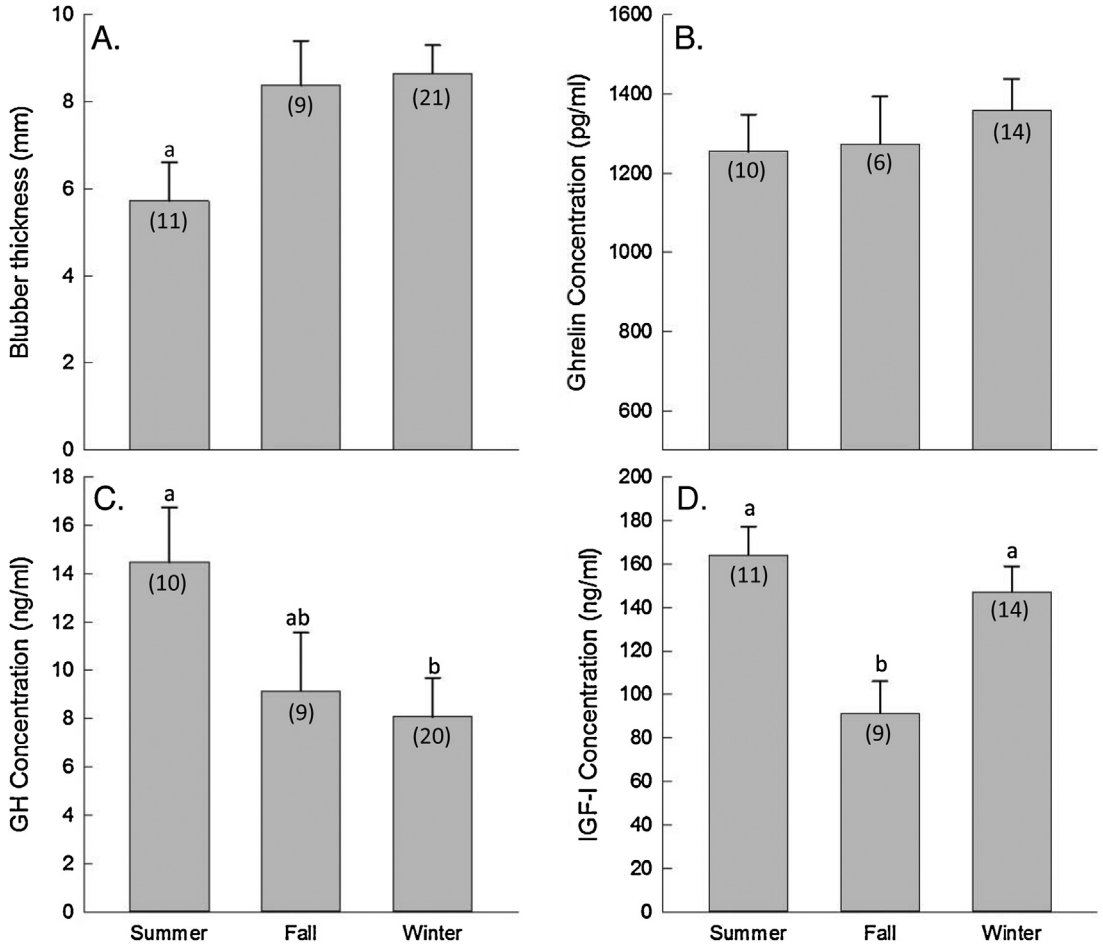

Fig. 1. (A–D) Pattern among seasons (summer, fall, winter) presented as mean ± SE in adult free-ranging manatees from 2005 to 2010. (A) Average blubber thickness (mm) (B) Average ghrelin concentrations (pg/ml) (C) Average GH concentrations (ng/ml) (D) Average IGF-I concentrations (ng/ml). Significant differences among seasons were presented as letters (a, b). Sample size is within the column of each season.

Breier, 1999). During energy deficiency or periods of low nutritional status, GH stimulates lipolysis and inhibits lipogenesis (Bergan et al., 2015). The mobilization of fat from adipose tissue, or lipolysis, provides energy for maintenance (Vijayakumar et al., 2010). When nutritional intake is adequate for energy requirements of maintenance, GH stimulates the production and release of IGF-I from the liver. Insulin-like growth factor-I regulates somatic growth, cell proliferation, differentiation, and apoptosis (Renaville et al., 2002). During normal physiological conditions IGF-I stimulates protein synthesis, bone formation, and glucose uptake (Etherton, 1991).

Modifications in nutrient intake or seasonal abiotic variables elicit an endocrine response resulting in alteration of growth rate and tissue-specific nutrient priorities (Gerrard and Grant, 2006). Mammals that exhibit seasonal changes in body composition are generally leaner during long photoperiod (summer, greater than 12 h of daylight) with greater adiposity during the short photoperiod (winter, less than 12 h of daylight; Florant and Healy, 2012; Webster et al., 1999; Zhang et al., 2012; Zhu et al., 2012; Zinn et al., 1986). Some mammals demonstrate weight gain, with increased adiposity, during a transitional period (fall) in preparation for the winter when food may be scarce, ambient temperature declines, and additional energy is needed for thermoregulation (Florant and Healy, 2012; Fuglei et al., 2004; Webster et al., 1999; Zhang et al., 2012; Zhu et al., 2012). During winter, some mammals minimize lean tissue accretion to conserve body protein (Nunes et al., 1998; Prestrud and Nilssen, 1992; Webster et al., 1999). Previous studies found that during winter ghrelin stimulates lipogenesis and food intake (Tschop et al., 2000). During fall GH was reduced to facilitate adipose accumulation in preparation for short photoperiod (Fuglei et al., 2004). In contrast, summer GH concentrations were increased to facilitate lean tissue accretion mediated by IGF-I (Rosenfeld and Hwa, 2009). In previous studies of ungulates, IGF-I concentrations were greater in summer compared with winter and were correlated with increased lean tissue accretion (Barenton et al., 1988; Comeau et al., 2008; Webster et al., 1996).

Most research on the role of metabolic hormones in the regulation of seasonal change in body composition has been examined in species living at middle to high latitudes that exhibit strong seasonal patterns in food intake and body composition (Fuglei et al., 2004). However, the physiological mechanism for seasonal body composition is unknown in sub-tropical or tropical species with less pronounced seasonal patterns. The free-ranging Florida manatee (*Trichechus manatus latirostris)*, a subtropical aquatic mammal, seasonally shift their habitat preference due to their narrow thermoneutral zone (Reynolds and Wilcox, 1986). Manatees tend to remain relatively inactive in warm water areas during cold air or ambient temperatures to reserve energy (Reynolds and Rommel, 1996; Reynolds and Wilcox, 1986). Therefore, manatees feed and forage less frequently resulting in negative energy balance and blubber loss (Gallivan et al., 1983; Reynolds, 2000). While seasonal changes in blubber thickness have been observed, the physiological mechanisms that contribute to this seasonal difference in body composition has not been explored in the Florida manatee. The primary objective of this research was to determine if manatee