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Morphometry and Body Mass Index to Assess Health Condition in Captive Population of Greek Tortoise, *Tesudo Greaca*

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Key Words: tortoise, body mass, chelonians, linear association, morphometryBody mass and morphometric data from a sample of captive Greek tortoise, <i>Testudo greaca</i> were collected to assess body condition based on linear association. Mass- length linear relationship differed between the sexes where mass-straight carapace length was strongly associated in males while mass- curved carapace length was strongly associated in females compared with other morphometric measures. Females were heavier, deeper and longer than males. Mass-length linear relationship was used to calculate the condition index log (M/m), where (M) is the observed mass and (m) is the predicted mass. The loss in body condition loss ranged between 1.5 to 13% and 2.7 to 15.9% in females and males, respectively. The mass-length technique is a useful means of evaluating health and growth in tortoises.
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1. INTRODUCTION

Many studies of animal ecology rely on nondestructive methods to estimate the body condition of different animal species in a population, relying on morphometric measures of body to calculate a body condition index (BCI) (Green, 2001; Stevenson and Woods, 2006; Peig and Green, 2009). This technique was widely used in a wide range of taxa, including reptiles (Bonnet and Naullea, 1994; Zwieg, 2003; Pasachnik et al., 2012), birds (Johnson et al., 1985; Alonso-Alvarez et al., 2002; Labocha and Hayes, 2012), mammals (Schulte-Hostedde et al., 2001; Trites and Jonger, 2000; Pitcher et al., 2000; Cattet and Obbard, 2005, Molnar et al., 2009), fish and amphibians (Brown and Murphy, 1991; Bister et al., 2000; Hansen and Nate, 2005; MacCraken and Stebbings, 2012).

Chelonians present a good candidate for using this technique to assess their body conditions both in the wild and in captivity because they are capable of withdrawing within the margins of the shell when threatened, making it difficult to collect samples for physiological parameters. A useful body condition index (BCI) for tortoises is log (M/m), where (M) is the observed body mass, (m) is the mass predicted from length (L) and (M/m) is relative mass (usually expressed as percentage)(Hailey, 2000; Willemsen and Hailey, 2002). Log (M/m) is equal to the residual from a regression of log (M) on log (L) that represents the condition index of the animal. An

individual with a positive residual is considered to be in a good body condition than an individual with negative residual (Dobson, 1992; Dobson and Michener, 1995; Guinet et al., 1998).

Despite the debate that accompanied the use of this technique, the residual approach is still considered the most reliable index of condition because it does not vary with body size (Jacob et al., 1996). Veterinary studies have also shown that body mass reflected in the condition index varies with health conditions (Jackson, 1980; Hailey, 2000). Therefore, the residual approach can be applicable to assess health status of animals.

The following research is an attempt to employ the residual approach on an endangered species of tortoise, the Greek , *Testudo greaca* under captive conditions. The main objectives of the study were to examine Specific morphometric measures that best fit linear relationship with body mass and can be used to estimate body condition index, differences between sexes in morphometric measures and in body condition indices that reflect the general health status of the population.

2. MATERIAL AND METHODS 2.1. SEXING AND WEIGHING

A total of 39 Greek tortoises were randomly selected from the captive population held in Kuwait zoo and transferred to the lab facility. The individuals were sexed by plastron concavity, shape of rear marginal scute and relative tail size according to the method adopted by Willesmen and Hailey (2002), Djordjevic et al.(2013). Individuals that measured less than 10 cm straight carapace length were excluded because sex separation was impossible. A Soehnle electronic balance was used to determine the mass (M) of each sex to the nearest one gm.

2.2. Morphometric Data

For each individual straight carapace length (SCL) was measured as the maximum horizontal length of the body in a normal flat position on the ground. Curved carapace length (CCL) was measured from tip of nutchal scute to the end of anal scute following the curvature of the carapace. Plastron length (PL) was measured from the tip of intergular to end of anal scute and plastron width was measured from middle of inframarginal shield left to right side. The carapace height (CH) was measured with taxonomic board in perpendicular way. All measurements were taken to the nearest 1mm with a measuring tape according to the methodology adopted by Bonnet et al. (2001), (Fig.1).

2.3. ANALYSIS

Comparison of a sample mean of all measures with expected value (CI=0, corresponding to μ = m) by using 2 sample T tests at 95% confidence limit was conducted. To assess the linear relationship between mass (M) and morphometric parameters and calculate body condition index, regression analysis was used for this purpose. Body condition index was estimated as the residual of body mass (M) on linear measures (Schulte-Hostedde et al., 2005). All statistical analysis were performed using minitab 13 and stata 12 softwares.

3. RESULTS

3.1. Sexing and body mass

The sample of Greek tortoise was separated by sex into 15 females and 24 males. There was a significant difference in mass between the two sexes with females being heavier than males (T test= 4.09, df= 34, p< 0.05, Table 1, Fig. 2).

3.2. Morphological measures

Table (1) shows different morphological measures in females and males. The linear relationship between body mass (M), straight carapace length (SCL), curved carapace length (CCL), carapace height (CH), plastron length (PL), and plastron width (PW) in males was highly significant ($F_{5,17}$ = 44.55, P< 0.001, R² (adj)= 0.90). By comparing the linear association of mass with all morphological measures it was evident that mass and straight carapace length established the strongest linear association ($F_{1,21}$ = 152.9, P< 0.001, R² (adj)= 0.88, Fig. 3). Analysis of mass on straight carapace length, therefore was used to generate the regression equation: log (m) = -4.34 + 3.25 log (SCL), where log (m) is the estimated mass from the equation.

All morphometric measures differed significantly between sexes (Table 1) where females were wider, deeper and more domed than males. The linear relationship between mass (M), straight carapace length (SCL), curved carapace length (CCL), Carapace height (CH), plastron length (PL) and plastron width (PW) in females was highly significant ($F_{5,9}$ = 18.7, p < 0.001, R² (adj)= 0.86). Among the linear association with other morphometric measures ccl was the best predictor of body mass ($F_{1,13} = 62.7$, P< 0.001, R² (adj) = 0.81, Fig. 4). Analysis of mass on curved carapace length therefore was used to generate the regression equation: log (m) = -3.29 + 2.7 log (CCL), where log (m) is the estimated mass from the equation.

3.3. Body condition index (BCI)

In females 47% of the population lost body condition as predicted from the regression equation (BCI with negative values). The loss in body weight ranged from 1.5 to 13% from the predicted mass value (Fig 5). Also in males 50% of the population lost body condition as predicted from the regression equation (BCI with negative values). The loss in body weight ranged from 2.8% to 15.9% from the predicted mass value (Fig. 6)

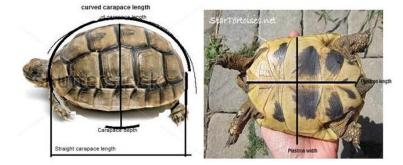


Fig. 1. Morphometric measures of the tortoise shell.

Sex	Mass (g)	Straight carapace length (mm)	Curved carapace length (mm)	Carapace height (mm)	Plastron length (mm)	Plastron width (mm)
Male	830.2	168.3	201.6	88.2	141.9	110.2
	±79.4	± 4.5	± 6.6	± 3.1	± 4.5	± 0.3
Female	1294*	187.8*	231.7*	103.7*	169.6*	123.6*
	± 81	± 4.2	± 5.2	± 1.9	± 3.8	± 3.02
T.value	4.09	3.2	3.58	4.6	4.6	3.2

Table 1. Comparison of morphometric parameters in the Greek tortoise (T. greaca) of both sexes.

Values are means ± Standard errors, P<0.05

SCL,(T= 3.2, df= 35 P < 0.05); CCL,(T= 3.58, df= 36, P < 0.05); CH,(T= 4.6, df=36, P < 0.05); PL, (T= 4.6, df=36, P < 0.05; PL, (T= 4.6, df=36, P < 0.05; PL, (T= 4.6

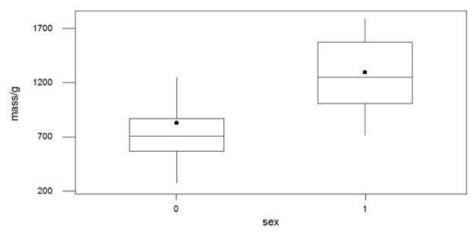


Fig. 2. Mass differences between males (0) and females (1).

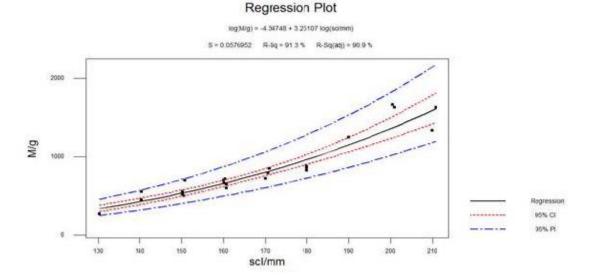


Fig.3. Relationship between mass (g) and straight carapace length(mm) in males.

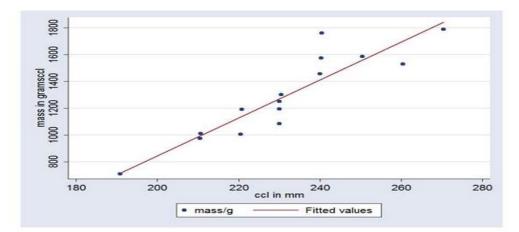
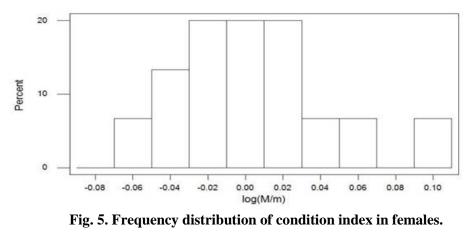


Fig. 4. Relationship between mass (g) and curved carapace length (mm) in females.($\log (m) = -3.29 + 2.7 \log CCL$).



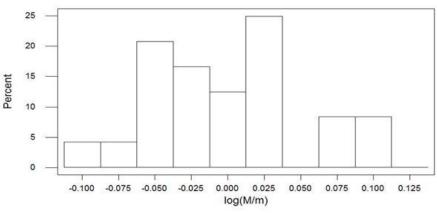


Fig. 6. Frequency distribution of condition index in males.

1. DISCUSSION

Body mass condition has been used with varying success in studies of chelonians. Spratt (1990) concluded that mass-length equations for wild populations of tortoises can be used for calculating body condition index in captive tortoises. The analysis of data on body mass and other morphological parameters between sexes showed that females were heavier, wider and deeper than males. Bonnet et al. (2001) in their study on *Testudo horsieldi* documented the presence of sexual dimorphism of carapace shape in the species. Females were significantly wider and higher than males at the same length. Sexual dimorphism in mass-length relationship may cause a problem if the BCI is calculated from regression pooling the sexes (Hailey, 2000). Therefore, the use of regression equation for each sex separately is appropriate to overcome the problem. The difference in body mass and carapace size in favor of females is probably related to egg production with large eggs relative to the size of shell (Kabigumila, 2002). Despite the findings of Kabigumila (2002) in relation to plastron length suggesting no difference between males and females of Pancake tortoise, other authors (Lambert, 1982) documented sexual dimorphism with respect to plastron length where male T.greaca were smaller, with prominent and acute scutes than females which agree with the findings in the current study.

There was a significant variation in BCI within sexes in this study where 47% and 50% of female individuals male were under weight, and respectively. Although BCI can act as an indicator for general health, interpretation of the results of BCI should be treated with caution. There are some factors that limit the usefulness of BCI in females before and after oviposition and between the activity seasons and just after hibernation (Willesmsen and Hailey, 2002). Body mass condition could reflect the level of hydration of a tortoise, the fullness of its gut, or the composition of body tissues, particularly changes in the mass of fat in the shell (Blaxter, 1989). The volume of a tortoise is relatively constant and the expansion of body tissues is constrained by the hard shell. Increased fat content, directly before hibernation may therefore be at the expense of other tissues of higher relative density, which would reduce the BCI, unlike most animals in which increased fat content is associated with higher body mass (Willesmsen and Hailey, 2002). This study was carried out at the beginning of the hibernation season in November when tortoises went into inactive state. This behavioral state might have had some effect on the BCI producing negative values in some individuals. Bidmon (2001) found that female tortoises lost on average 5.5% of its body mass during hibernation while male tortoises lost 11%. Regardless of the real factors generating low body condition in some of the studied individuals the technique of mass-length relationship to assess body condition is an acceptable method for health and growth evaluation in tortoises. In conclusion the body mass-length technique could be a practical noninvasive method for assessing body condition and general health status for our captive tortoises where 40 to 50% of the population was under-weight.

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