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Easy life of males? Indirect evidence that growth is easier than egg production in mangrove-dwelling monitor lizards (*Varanus indicus*)

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Abstract. In male-larger species of animals, males typically continue to grow after the age of female sexual maturation has been reached. Consequently, a switch of energy allocation occurs as the female investment from growth is shifted into egg production. We focus on the transitional period when both sexes heavily invest into anabolic processes; males invest in the development of body tissues while females predominantly invest in egg production. In captive mangrove-dwelling monitor lizards, we found that relative food intakes as well as quantitative estimates of anabolic processes (relative growth and egg production rates) are fairly comparable between the sexes. In spite of this biochemical clinical values and body condition indices revealed sex differences suggesting costs of reproduction in females. These results clearly illustrate that growth and egg production still substantially differ in associated physiological costs. This may be attributed to qualitative requirements (nutrients, minerals, etc.) of these processes. Our results correspond well with the higher susceptibility and mortality rates of females than males in many lizard species in captivity.

Keywords. Bimaturism, biochemical clinical values, growth rate, reproductive investment, sexual size dimorphism, Varanidae

INTRODUCTION

In most animals, fundamental differences between the biological roles of males and females arise as an inherent consequence of sexuality (Trivers, 1972, 2002). Ova are considerably larger gametes than sperms and consequently, the value of minimal expenditure per offspring is much higher in females than in conspecific males (Trivers, 1972, 2002). This potential inequality in reproductive investment between the sexes responsible for sexual selection is especially highlighted in animal taxa producing large offspring, e.g., terrestrial vertebrates (Clutton-Brock, 1988). In these animals, females are imprisoned to pay heavy metabolic costs to produce eggshells and yolk (or equivalent nutrition for the embryos in the case of viviparous taxa). In contrast, male investment (except in species with paternal care) is more variable and a considerable part of it is allocated to activities associated with intrasexual competition, e.g., territory defence, male combat, and mate guarding (Darwin, 1871; Andersson, 1994).

In reptiles, the females' cost of reproduction was demonstrated repeatedly (Itonaga et al., 2012; Vitousek et al., 2010; for a review see Schwarzkopf, 1994). Fitness of an individual depends, besides other factors (e. g., behavioural traits), on the decisions determining the allocation of available resources to particular life-history traits. Consecutive hierarchical allocation of female resources was clearly demonstrated in *Paroedura picta,* a small male-larger gecko with an invariant clutch size. Its females' investment is first allocated to growth, second to reproduction and finally to fat storage (Kubička

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and Kratochvíl, 2009). Also, in other male-larger lizards, females usually slow their growth after maturation and allocate further investments preferably to reproduction. In contrast, males of the same age continue to grow intensively as larger male size is usually attained by prolongation of the growth period, resulting in sexual bimaturism (Stamps and Krishnan, 1997; Frynta et al., 2010). Supposing that other energetic expenses (e.g., basal metabolism, locomotion, etc.) are comparable between the sexes and that animals allocate all remaining energy to anabolic processes, these should also be comparable between males and females during this transitive period. Nevertheless, males spend the entire energy budget into the building of body tissues, while females allocate the resources saved in the expense of growth into the production of eggs. Investments into the production of eggs are constrained by the availability of minerals as calcium (de Buffrénil and Francillon-Vieillot, 2001; Kratochvíl and Frynta, 2006). Thus, we may expect qualitative differences between the production of eggs and body tissues. We hypothesise that it is not the energetic expenses, but rather the qualitative differences that may determine the relative costs of male and female types of investment.

The period of ontogeny when both sexes are pressed to maximize anabolic processes, but differ in their preferred allocation, provides a promising model. Males allocate the energy into the growth of their bodies, while females into the egg production. We can compare their investments in terms of produced biomass, required energy (both scaled to body mass and/or metabolism), and associated health costs.

The mangrove-dwelling monitor, *Varanus indicus* (Daudin, 1802) belongs to the subgenus *Euprepiosaurus* (cf. Ast, 2001; Böhme, 2003). This group consists mainly of closely related forms that were recently split from *V. indicus* sensu stricto (Ziegler et al., 2007; Koch et al., 2009). *Varanus indicus* is capable of indeterminate (de Buffrénil et al., 2004) and rapid growth (cf. cortical vascularization of long bones; de Buffrénil et al., 2008). The mangrove-dwelling monitor lizard is among the most sexually dimorphic monitor lizards of the family Varanidae (Frýdlová and Frynta, 2010). This sexual dimorphism especially concerns body size, while shape differences between the sexes are rather small (Frýdlová et al., 2011). Under natural conditions, males are about 2.66 times heavier (mean body mass = 1287 g) than females (484 g; Wikramanayake and Dryden, 1988). The degree of sexual size dimorphism (SSD) computed from asymptotic body weights estimated in captivity was as high as 2.81 (4594 g in males versus 1635g in females; Frynta et al., 2010). Similarly to other species of monitor lizards (Auffenberg, 1981; Lemm et al., 2004), *V.indicus* is not only large, but also a surprisingly fast-growing animal. The sexes differ both in their asymptotic body sizes and the timing of growth (corresponding to somewhat delayed inflection points of the logistic growth curves of males compared to that of females). In contrast, maximum growth rate (in the exponential phase of the growth curves) are almost the same in males and females. Nevertheless, the absolute growth rates, and partially also relative growth rates in the next phase of growth (at the age of 500-1000 days), continue to be high in males, while females gradually stop to grow and switch the energy investment into egg production (Frynta et al., 2010).

In this paper, we analysed data concerning males and females of *V.indicus* during the phase when males still continue to grow intensively while females retard their growth in favour of reproduction. We estimated growth rates, egg production and food intake. The data concerning the investment of the sexes into anabolic processes were compared with chemical clinical values collected at the same period to characterize the health and nutritional status of the examined animals. This allowed us to test the hypotheses that (1) the investment into anabolic processes is comparable in both sexes; (2) female investment in egg production is more costly (in terms of health parameters) than corresponding male investment into their own body tissues.

MATERIAL AND METHODS

The experimental animals were 34 individuals (21 males and 13 females) of mangrove-dwelling monitors, *Varanus indicus*, reared at the Prague zoo. They were siblings belonging to four subsequent clutches hatched in November 2006, January, February and June 2007. The animals were housed individually in terrariums gradually changed for larger ones to satisfy the needs of the growing animals (ending with terrariums 145 x 100 x 130 cm large). The ambient temperature in the breeding room varied from 28 to 30 °C with hotter basking sites. Chicken livers or necks were provided weekly (ad libitum), supplemented with vitamins and minerals (Nutri Mix, Trouw Nutrition Biofaktory, Ltd.). AD_3 and E vitamins (PHARMAGAL s.r.o.) were provided monthly.

In order to provide unambiguous sex determination (cf. Schildger et al., 1999; Horn, 2004) and to assess the reproductive status, we employed ultrasonographic imaging. On 15 February 2009, we performed ultrasonographic examination of abdominal cavities using a linear electronic 7.5 – 14 MHz probe (Sonix OP, Kanada) with the animal lying in dorsal recumbence. All abdominal ultrasound scans were performed systematically in a standardized fashion from cranial to caudal direction in both sections (longitudinal and sagital) and were performed by one examiner (J.H.). In females, follicle size was measured to assess the stage of ovarian cycle that may affect their physiological state. Ultrasonic imaging revealed that matured follicles or ova

were already present at the age of 506 days (Frynta et al., 2010). The first clutch of unfertilized eggs was recorded at the age of 578 days (Frýdlová, pers. comm.). Ultrasonography revealed sexual maturation in experimental animals of both sexes and this conclusion was further confirmed by observation of mating during the experimental encounters. Since the age of five months, we allowed short-time encounters between the sexes to test signs of sexual behaviour and recorded the first mating attempt of a male at the age of 522 days.

The experimental animals were regularly weighed every two weeks (before feeding) and snout-vent length was measured every three months. Increments of body weight (body weight as assessed at a given moment minus that at a previous weighting) were calculated for each experimental animal and each two-week period within the range of 500-1000 days. These were divided by the exact length of the period (in days) and scaled to body weight at the beginning of the period. The observed increment represents the mass of produced tissues that should be roughly proportional to the allocation of energy to anabolic processes.

In females, the partial increments were affected by ovarian cycles involving gradual increase of total body weight during the growth of ova followed by an abrupt body weight loss at egg deposition. However, these temporal effects affecting the variation of the increments had no effect on mean values, because increases were fully compensated by weight losses when averaging weight increments assessed during the time frame exceeding the length of the ovarian cycle (two months).

The amount of available body tissue strongly determines the quantity of metabolic processes. Thus, we can reasonably expect that the body weight at the beginning of the period roughly reflects the metabolic (both anabolic and catabolic) capacity of the individual and thus, it is a reliable variable for the standardization of body weight increments. Because the exponent of the allometric relationship between metabolic rate and body weight is usually (Kleiber´s law; Schmidt-Nielsen, 1984) closer to 0.75 than to 1 (corresponding to isometry), we alternatively scaled the body weight increments to the initial body weight^0.75.

The egg production of each female was monitored throughout the study and each egg was weighed prior to its placement into the incubator. Because the intervals between clutches were fairly regular, lasting about two months, we were well informed about the approximate timing of egg laying. During this period, the behaviour (e.g., digging) of the females was observed to prevent egg cannibalization and/or damage.

Food intake was measured as the amount of food that disappeared from the feeder (plastic box with surplus food corresponding to about 30% of the body weight) during 24 hours. It was assessed once on 19-20 January 2009 (liver diet; further referred to as period 1) and repeatedly 4-5 times from 2 March to 6 July 2009 (chicken neck consumption; further referred to as period 2). The age of the experimental animals at the time of food intake assessment varied within the range of 590-810 and 632-978 for periods 1 and 2, respectively. Food conversion rate was calculated as food consumption (in grams) divided by body weight increment assessed in a given two-week period (in grams).

The health condition of the experimental animals was monitored regularly. On the 14 December 2008, 15 February 2009 and 25 April 2009, we performed complex veterinary surveys including the collection of blood samples for the assessment of biochemical clinical values. Blood samples were taken from the ventral caudal tail vein. The blood was sampled by direct puncture with a 20G needle and a 2 ml heparinized syringe. About 0.8 to 1.5 ml of whole blood was aspirated and directly sent for biochemical analysis, blood smear and blood count. For biochemical testing whole blood was analysed using the dry chemistry system Spotchem EZ (Medista, CZ). We measured 8 biochemistry values: concentration of albumin (Alb), transferins = other protein (Trans; calculated as Total protein - Alb), total bilirubin (T - Bil), glucose (Glu), calcium (Ca), inorganic phosphorus (IP), uric acid (UA), alkaline phosphatase (ALP).

The concentrations of proteins (Alb, Trans) are usually treated as markers of nutritional status and body reserves; glucose (Glu) levels reflect the actually available energy; relationship between the concentrations of main minerals (Ca, IP) reflects the calcium balance; and the elevated concentrations of excretion products of metabolism (T-Bil, UA) and ALP are treated as markers of uncompensated metabolic load (Campbell, 2006; Diethelm and Stein, 2006).

Scaled mass index (SMI) reflecting body condition was calculated after Peig and Green (2009, 2010): $SMI = M_i * (L_o/L_i)^{bSMA}$ (M_i = body mass; L_i = body length, i.e., SVL in our dataset; L_o = arbitrary value of L_i, e.g., close to arithmetic mean values of body length in study population; bSMA is a slope of reduced major axis of body mass versus body length allometric relationship). Empirical RMA slope calculated from our data was 3.3225, L_o was set to 400 mm). The RMA slope was calculated from entire data points (pseudoreplicates) available for the range of 500-1000 days. Prior to this analysis, we confirmed that the slope of the allometric relationship of body weight and SVL did not differ between the sexes. For this purpose, we run a marginal GLS model (see below) accounting for repeated measures that revealed no effect of SVL*sex interaction ($F = 0.03$ and $P = 0.8557$). The values of SMI were highly correlated ($r = 0.957$) with those of Fulton's index of condition which is calculated as $K = M/L^3$. Thus, both these indices of body condition were mutually interchangeable and, following Peig and Green (2010), we selected SMI for further analyses of body condition at the time of blood sampling.

We used STATISTICA Analysis System (version 6.0) for most calculations. The marginal models including accounting for repeated measures was performed in R version 2.10.0 (R Development Core Team 2009) using GLS function as implemented in the nlme package. Symmetric correlation structure was selected for these analyses.

The experiments were performed in accordance with the Czech law implementing all corresponding European Union regulations and were approved by the institutional animal care and use committee (5530/2008-30, 8388/2011-30).

RESULTS

Investment to body growth and egg production

Median values for body size of males and females during the study period were: 432.4 mm (n = 21, range

333.0 – 550.7 mm) and 369.6 mm (n = 13, range 308.3 – 433.2 mm), respectively. Weight increments per day scaled to body weight^0.75 were examined using marginal models accounting for repeated measuring of the same individual (pseudoreplications). First, we included sex, log-transformed age and their interaction as factors. The effect of the interaction was not significant ($F_{(1,985)}$ = 0.34, P = 0.5606) which suggested homogeneity of slopes and allowed us to remove the interaction from the model (this decision was confirmed by the log-likelihood ratio test at α = 0.05). The reduced model revealed that weight increments per day scaled to body weight^0.75 were higher in males than in females ($F_{(1,986)}$ = 103.47, P < 0.0001, coefficient for males = 0.00888, SE = 0.000873) and that they decreased sharply with log-transformed age ($F_{(1,986)}$ = 47.90, P < 0.0001, coefficient = - 0.02004, SE = 0.00275; Fig. 1). Thus, male investments into the production of new body tissues is higher than that of females of the same initial body mass and this difference may be interpreted as the anabolic capacity saved by the female in expense of growth.

In order to estimate the amount of the anabolic capacity saved during a single ovarian cycle by a typical female, we performed the following calculations. Per two month period (= the typical inter-clutch interval) and initial female body weight^0.75, a female typically saves the amount of reserves needed to build 0.533 grams of its body (= 0.00888[grams per day]*60[days]). Considering that at the age of 500-1000 days, the mean female body mass was 1071 g (n = 368 pseudoreplicates), which corresponds to 187.7 in terms of weight^0.75, a female saved 99.8 g of its body weight (= 0.533*187.7) that may be invested elsewhere (e.g., into egg production). Because mean weight of eggs produced by these females was 25.21 g, this represents a rough equivalent of four eggs (99.8/25.21 = 3.96 eggs).

We alternatively performed this calculation based on the relationship between body weight increment and untransformed body weight of the female. The difference between the intercepts of this relationship in males and females (i.e., the coefficient for the male sex) was 0.066145. Thus, per two month period and per gram of its initial body weight, a female typically saves the amount of reserves needed to build 0.066145 grams of her body (0.066145 = 0.001102409 [grams per day]*60[days]). A female of mean body weight of 1071 g thus saved the investment of 70.8413 g (= 0.066145*1071), which corresponds to 2.81 eggs.

During the study period, we recorded 28 clutches (n females = 10) consisting of 1-10 eggs. The mean clutch size and mean egg weight were 3.96 eggs and 25.21 g, respectively.

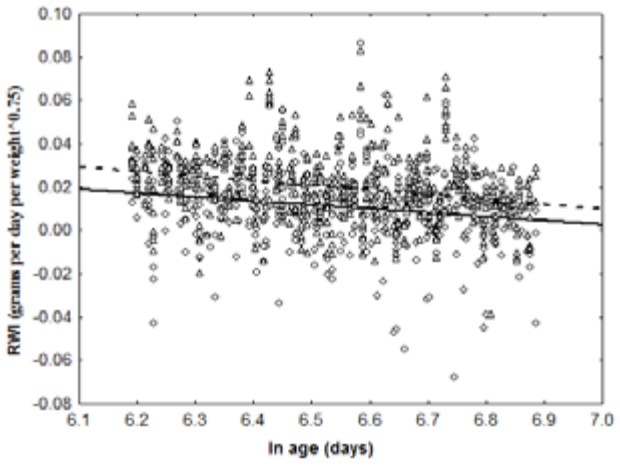

Fig. 1. Plot of the relationship between relative weight increments (RWI, grams per day per weight^0.75) and log-transformed age (ln age) in captive mangrove-dwelling monitor lizards at the age of 500-1000 days. Lines indicate ordinary least-square (OLS) regressions computed separately for males and females. The equations of these regressions are as follows: females: y = 0.128960606 - 0.0180518898*x (r = -0.1788, P = 0.0006); males: y = 0.160058468 - 0.0214532419*x (r = -0.2630, P < 0.0001). Triangles are males, circles are females.

Food intake, its scaling and rates of food conversion into body mass

We found no sex differences in food intake per body weight (period 1: means = 0.599 and 0.611 in males and females, respectively; $n_{(males)}$ = 21, $n_{(females)}$ = 13; t = -1.09, P = 0.2841; period 2: 0.405 and 0.426; t = -0.79, P = 0.4351). When food intake was scaled to body weight^0.75, these values appeared somewhat higher in males than in females. Nevertheless, this difference was statistically significant in period 2 only (period 1: 2.690 and 2.405; t = 1.694, P = 0.1000; period 2: 1.812 and 1.436; t = 3.40, P = 0.0018).

In males, allometric relationships between food intake and body weight were clear (r = 0.802 and 0.554; Ps<0.0001), the intercepts (A) and slopes (B) of these relationships were A = 1.3271, B = 0.702 (SE_B = 0.120, n = 21) and A = 1.2377, B = 0.658 (SE_B = 0.109, $n_{(pseudoreplicates)}$ = 84), in the first and second period, respectively. The confidence intervals of the slopes overlap the theoretical values 0.75. In females, this allometric relationship was significant in the first period (A = 2.1266, B = 0.568, SE_B = 0.212, n = 13, r = 0.631, P = 0.0209), but not in the second period (A = 8.0803, B = -0.340, SE_B = 0.299, $n_{(pseudoreplicates)}$ = 53, r = -0.157, P = 0.2608). The homogeneity of slopes test revealed sexual differences in allometric slopes

during the second ($F_{(1,133)}$ = 12.53, P = 0.0006; $F_{(1, 119.95)}$ = 16.04, P<0.001), but not the first period ($F_{(1,30)}$ =0.27, P = 0.6096).

In the first period, the mean food conversion rate was 18.2% in males and 13.8% in females (Mann-Whitney test, $n_{(males)}$ = 21, $n_{(females)}$ = 13, z = 0.62, P = 0.5351). In the second period, the food conversion rate was 16.8% in males and 1.3% in females, and this difference (computed from mean values for each individual) was significant (Mann-Whitney test, $n_{(males)}$ = 21, $n_{(females)}$ = 13, z = 3.07, P = 0.0022).

Biochemical values and body condition index

Means and other descriptive statistics of non-transformed biochemical values are given in Table 1. We performed PCA in order to examine the correlation structure among the studied biochemical values to reduce the number of examined traits. PC1 and PC2 accounted for 34.1% and 21.3% of variation in the original log-transformed variables, respectively (see Fig. 2 for loadings). PC1 is correlated positively with bilirubin, phosphor and uric acid (i.e. metabolic residues) and negatively with albumin, glucose and calcium (i.e., the actually available sources of amino acids, energy and minerals). Thus, it may be interpreted as a measure of body reserves deficit. PC2 scores increase with values of albumin, calcium, transferins and uric acid.

PC scores were further examined to assess the effects of sex; PC1 scores were smaller in males than in females and this difference was significant (marginal GLS: $F_{(1,88)}$ = 12.98, P = 0.0005, intercept = 0.6327297, SE = 0.2059295, coefficient for males = - 0.8899365, SE = 0.2470415). No significant sex differences were found in PC2 scores (marginal GLS: $F_{(1,88)}$ = 1.96, P = 0.1648). In females, PCs were apparently not associated with follicle size (PC1: n = 10; r = - 0.133, P = 0.7139; PC2: n = 10; r = -0.209, P = 0.5624), nor the presence/absence of egg laying during the week preceding blood sampling ($n_{(pseudoreplicates)}$: absent = 19, present = 8; PC1: t = 1.20, P = 0.2426; PC2: t = 1.14, P = 0.2664).

The scaled mass index (SMI) was slightly, but significantly higher ($F_{(1,83.54)}$ = 6.24, P = 0.014) in males (mean = 1603; SE = 34.99) than in females (mean = 1498; SE = 23.07). Nevertheless, the relationship between this body condition index and PC1 of biochemical values was not tight and interacted with sex ($F_{(1,61.5)}$ = 6.84, P = 0.011; Fig. 3).

Table 1. Means, standard deviations (SD), medians, percentiles (5 and 95%) of the clinical chemical values in the males and females of the mangrove-dwelling monitor lizards (*Varanus indicus*). The results of the testing of the sex differences by repeated measures GLMMS (F value, degrees of freedom and P value) are provided in the right column. Abbreviations: M – male, F – female, n – number of pseudoreplicates (blood samples), Alb – albumin, Trans - transferins (= other protein calculated as Total protein - Alb), T-Bil - total bilirubin, Glu - glucose, Ca - calcium, IP - inorganic phosphorus, Ca/IP – ratio of calcium and inorganic phosphorus, UA - uric acid, ALP - alkaline phosphatase. Significant comparisons are indicated by asterisks.

	Sex	n	Mean	SD	Median	5%	95%	F (df) P
Alb	M	63	20.59	3.87	21.0	13.0	27.0	1.77 (1, 74.4)
(g/L)	F	36	19.28	4.37	19.0	12.0	28.0	0.188
Trans	M	63	47.19	7.03	46.0	38.0	58.0	0.24 (1, 60.1)
(g/L)	F	36	48.19	12.16	46.0	31.0	76.0	0.624
T-Bil	M	63	8.70	4.39	7.0	5.0	17.0	4.64 (1, 80.0)
(μmol/L)	F	36	12.75	6.04	12.0	4.0	23.0	0.034*
Glu	M	63	6.55	1.09	6.5	5.2	8.4	12.30 (1, 69.3)
(mmol/L)	F	36	6.13	1.50	6.0	4.2	8.9	0.001*
Ca	M	63	2.84	0.32	2.9	2.2	3.3	1.20 (1, 77.7)
(mmol/L)	F	36	2.81	0.46	2.8	2.1	3.7	0.277
IP	M	63	1.67	0.34	1.6	1.2	2.3	7.27 (1, 77.9)
(mmol/L)	F	36	2.02	0.80	1.9	1.2	4.5	0.009*
Ca/IP	M	63	1.78	0.42	1.7	1.1	2.4	-
(mmol/L)	F	36	1.55	0.52	1.5	0.7	2.4	-
UA	M	63	194.75	59.49	188.0	112.0	322.0	50.15 (1, 64.49)
(μmol/L)	F	36	309.50	97.11	302.5	180.0	528.0	<0.001*
ALP	M	63	6.41	1.20	6.5	4.2	8.4	<0.01 (1, 86.8)
(μkat/L)	F	36	6.27	1.33	6.2	4.1	8.2	0.961

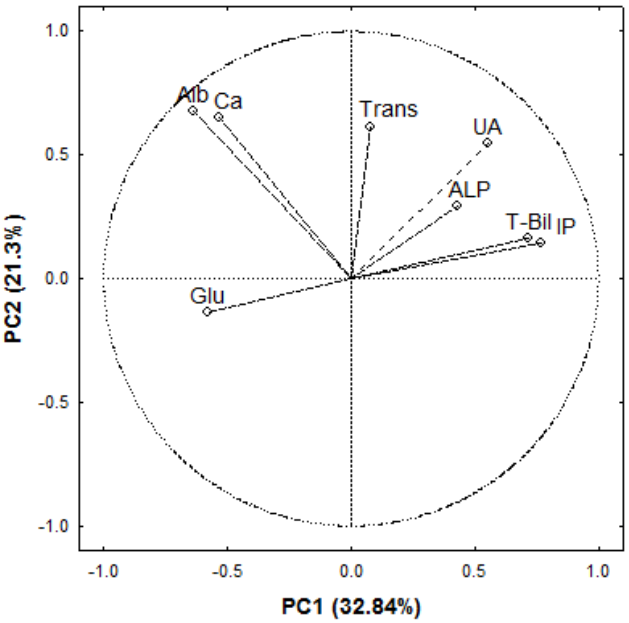

Fig. 2. Variation structure of the clinical chemical values in captive mangrove-dwelling monitor lizards visualized by loadings of the principal components (PC1 and PC2). Abbreviations: Alb – albumin, Trans - transferins (= other protein calculated as Total protein - Alb), T-Bil - total bilirubin, Glu - glucose, Ca - calcium, IP - inorganic phosphorus, Ca/IP – ratio of calcium and inorganic phosphorus, UA - uric acid, ALP - alkaline phosphatase.

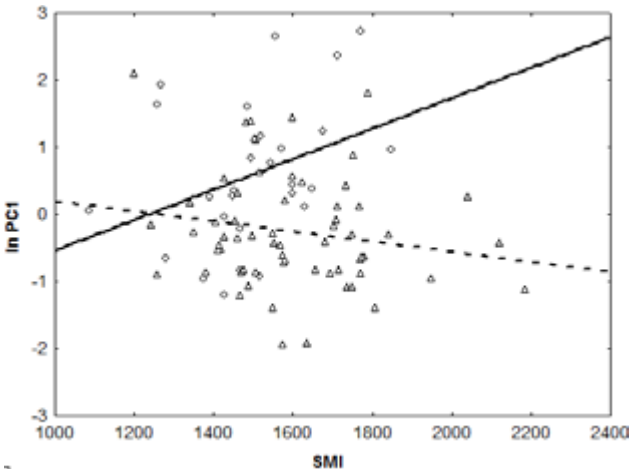

Fig. 3. The relationship between the principal component 1 of the clinical chemical values (PC1) and the scaled mass index (SMI) of the body condition at the time of blood sample collection in the captive mangrove-dwelling monitor lizards (*Varanus indicus*). Triangles are males, circles are females.

DISCUSSION

Investment to body growth and egg production

Authors studying maternal investment in lizards adopted the concept distinguishing between income and capital breeders (Schwarzkopf, 1994 and references herein; for concept see Drent and Daan, 1980). Income breeders produce eggs exclusively in the expense of actual food intake. In contrast, capital breeders are able to accumulate and store fat reserves for a long time period and invest them all at once. Nevertheless, these strategies represent a continual scale rather than a strict dichotomy. It was demonstrated in the scink *Eulamprus tympanum* that litter size depends on fat reserves while new-borns' body size depends on the actual energetic income (Doughty and Shine, 1998). Recently, Warner et al. (2008) measured isotope contents in Australian jacky dragons (*Amphibolurus muricatus*) and demonstrated that the energy allocation strategy used for reproduction differs sharply even among the egg components. Egg lipids follow the capital breeding strategy, while egg proteins follow both the income and capital ones. This may be explained by easier storing of fat compared to proteins.

As many other tropical lizards, mangrove-dwelling monitor lizards are predominantly income breeders. This assumption was supported by their growth patterns (Frynta et al., 2010; this study). In order to quantify sex differences in the quantity of anabolic processes during the examined period (age 500-1000 days), we measured the gains of body weight as well as the weight of produced eggs. In *V. indicus*, mature females produce clutches in regular intervals of approximately two months. Our females produced about 100 g of eggs per single ovarian cycle. This value fits well in our estimates of the mean difference between the body weight gains of males and females per two month period (rescaled to mean female body size in the study period). It equals to 99.8 g or 70.8 g, for allometric models with slopes 0.75 and 1, respectively. Thus, the egg production is realized mainly in the expense of saved metabolic reserves that are devoted to growth in males. Nevertheless, the energetic costs associated with the production of the body tissue may be somewhat lower than those associated with the production of the same amount of egg mass. Although the energetic value of eggs expressed in calories per dry weight is only slightly higher to that of body tissues (Congdon et al., 1978), we are aware that the differences in water content between these substances may affect this comparison considerably.

The temporal coincidence and quantitative correspondence between growth reduction and beginning of egg laying provides no conclusive information about the direction of the putative causal relationship between these two events. To solve this, experimental manipula-

tions with egg size and/or number (Landwer, 1994; Sinervo, 1994), castration (Cox et al., 2010) and food intake (Kubička and Kratochvíl, 2009) are required.

Food intake, its scaling, and rate of food conversion into body mass

It should be considered that our experimental study uses captive-bred animals. Ad libitum feeding, the absence of predation pressure, stability of environmental condition and the lack of intraspecific agonistic interactions would affect the experimental animals considerably. Compared to wild animals, the captive ones are capable to invest more energy to the body growth and reproduction (Mendyk, 2011). Retes and Bennett (2001) notify extreme cases of multiclutching (14 consecutive clutches in a 14-month period) in females of *V. glauerti* when treated under ad libitum conditions.

In our study, the relative amount of food consumed by males was roughly equal or even somewhat higher than that consumed by conspecific females. As food was provided ad libitum, the female metabolic loads did not substantially exceed those of the males during the examined period. Although males were larger than females, this conclusion remained substantially unchanged irrespective of scaling the food intake alternatively to body weight or predicted baseline metabolism (estimated as body weight^0.75). There are theoretical arguments supporting each of these scaling strategies (Sibly et al., 2012) and our empirical data suggest that the exponents of the allometric relationship between food intake and body weight support the exponent 0.75 when OLS regression (supposing that the error in body weight is negligible) is considered. However, the RMA regression model (considering errors in both axes) would result in somewhat higher slopes close to 1. Moreover, during period 2, there was no clear association between food intake and body weight in females, probably as a result of the great variation in both the food consumption and the actual body weight in egg-laying females.

We found that the rates of food conversion into body mass were initially (period 1) almost the same in males and females. Nevertheless, in period 2, these rates dramatically decreased due to the reduction of female investment into growth and the beginning of egg production.

Biochemical values and body condition index

The clinical chemical values reported by previous authors in other species of monitor lizards (*V. varius*: Jessop et al., 2011; *V. komodoensis*: Gillespie et al., 2000; Lemm et al., 2004; *V. dumerilii*: Bertelsen et al., 2007) are generally comparable to those of this paper. The differences are small enough to be attributed to differences in the methods of the assessment and small sample sizes.

We found that during the examined ontogenetic period, males of *V.indicus* exhibited higher values of glucose and lower values of urea, bilirubin, and phosphor, than females. We interpret these differences as indirect evidence that the body reserves and health condition of females were more limited than those of males. This corresponds well with the sex differences in the scaled mass index suggesting better body condition of males. Because the relative food intake, as well as the rates of the produced body/egg mass were at least comparable between the sexes (see above), the impaired body reserves and/or health condition of females cannot be explained by the increased loads of metabolic and/or anabolic processes in this sex. Thus, qualitative rather than quantitative differences between the loads associated with the production of eggs and body mass should be considered. This may lead to the conclusion that the cost of female duties is heavier than that of male ones. Although fairly intuitive, such evidence was not previously reported, at least not in large carnivorous lizards. However, temporal instability of calcium, phosphor and magnesium levels was clearly demonstrated in the reproducing females of Komodo dragon (*V. komodoensis*; Lemm et al., 2004). Recently, Laver et al. (2012) revealed substantially higher mortality rates in females than in males of wild *V. komodoensis* and attributed this finding to the heavy costs of reproduction in females. This further biases the operational sex ratio in favour of males and intensifies the strength of the sexual selection. According to life-history models (Charnov, 2011; Charnov and Warne, 2011), increased mortality should further reduce the investment into growth, health and longevity in females.

In conclusion, the empirical experience of breeders that females are a more susceptible and gentle sex was supported even in the model lizard with quantitatively comparable metabolic and anabolic rates in the sexes. This may be interpreted as a demonstration of a load associated with female life-history. Obviously, this load results from qualitative rather than quantitative differences between the metabolic duties of the sexes. This study was performed to stimulate further attention to these phenomena. Nevertheless, as any observational study, it is limited to correlative results and their interpretations. Thus, there is an urgent call for manipulative experiments enabling to uncover the causative relationships and providing independent controls (e.g., for sex differences in the absence of growth and/or reproduction).

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