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The influence of the incubation environment and parental care on Tokay gecko (*Gekko gecko*, Gekkonidae) development in captivity

Birgit Szabo [a, b] *

[a] Division of Behavioural Ecology, Institute of Ecology and Evolution, University of Bern, Bern, Switzerland

[b] Centre for Research on Ecology, Cognition and Behaviour of Birds, University of Gent, Belgium

*Correspond to: Birgit Szabo, Sint-Pietersnieuwstraat 33, 9000 Gent, Belgium; email: birgit.szabo@gmx.at,

ORCID BS: 0000-0002-3226-8621

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Abstract

Parental effects, non-genetic effects that parents exert on their offspring, can result in adaptive phenotypes that ensure offspring survival. In captivity, reptiles often experience unnatural incubation and rearing conditions due to limited nest site choices, which can negatively impact individual development and welfare. Incubation temperature and humidity are especially critical for reptile development, influencing factors such as sex determination, growth, and morphology. However, the conditions experienced after hatching, such as social housing, are far less often considered. Here, I studied the effects of both the incubation environment (temperature and humidity) and social rearing compared to isolation rearing on the development of captive bred tokay geckos (*Gekko gecko*). I find that both temperature and humidity affect hatchling sex and size but only temperature affected incubation duration. Furthermore, females selected nest sites with higher temperatures. After hatching, rearing group size did not affect Snout vent length or growth, but body condition was higher in offspring raised alone and in smaller family groups compared to larger family groups. My findings confirm temperature dependent sex determination in tokay geckos and a role for humidity in lizard development. Importantly, being raised in larger social groups led to a reduction in body condition across the first six months of life, which was not reflected in SVL alone. My results suggest that despite ample food available, competition within family groups does reduce offspring body condition. Therefore, care should be taken when raising tokay geckos in captivity to provide nest site options and enough food and shelter to ensure optimal development of all offspring.

Keywords: Captive rearing, enrichment, husbandry, reptile, social housing, welfare

Introduction

Parental effects are non-genetic effects that parents may exert on their offspring through developmental plasticity (Uller 2008). Parental effects can result in adaptive phenotypes that ensure offspring survival in the wild if the parental environment successfully predicts the future environment that offspring will encounter (Lindström 1999; Uller 2008). Both the pre- and postnatal environment that parents choose or create are sources that cause differential development in offspring (e.g. Bestion et al. 2014; Monteith et al. 2012). In captivity, reptiles often only have limited choice as to the environment their offspring will incubate in and juveniles are often raised in isolation to ensure healthy growth and easy health monitoring (Burghardt and Layne-Colon 2023). How deviations from natural conditions before and after hatching or birth affect the development of captive individuals, and in turn, their welfare and suitability for reintroduction programs, remains poorly understood (Crates et al. 2023).

One of the most important factors shaping development in reptiles is incubation temperature. Suboptimal thermal incubation environments, either too low or too high, can influence incubation time and lead to reduced survival as well as body deformities at birth and after hatching (Burghardt and Layne-Colon 2023; Noble et al. 2018). Even after birth or hatching, suboptimal incubation temperatures can have long-term effects on morphology and growth rate for up to one year (Noble et al. 2018). Importantly, in some lizard taxa such as geckos, agamas and lacertids, incubation temperature determines the sex of the hatching individuals (Burghardt and Layne-Colon 2023; Gutzke and Crews 1988). Apart from temperature, incubation humidity has also been shown to influence development in egg laying species with the potential to have similar effects on morphology and survival at suboptimal levels, too wet or dry, as incubation temperature (Bell et al. 2025). Incubation mimicking natural condition in captivity has been shown to improve development (Hall and Warner 2020), produce important phenotypic variation (Shine et al. 1997a) and should, therefore, be considered important in captive breeding.

In mammals and birds, it is generally assumed that natural rearing by parents or with siblings is more beneficial for the development of offspring compared to hand rearing by humans (Burghardt and Layne-Colon 2023). In lizards, parental care after hatching is uncommon but not absent and should, therefore, be considered when raising individuals of such social species in captivity (Burghardt and Layne-Colon 2023). On the one hand, rearing in isolation can negatively influence development of behaviour such as in juvenile veiled chameleons (*Chamaeleo calyptratus*) which showed deficits in social and feeding behaviour when raised alone compared to group rearing (Ballen et al. 2014). However, on the other hand, it should be noted that social housing might also have negative effects. Housing lizards in groups of siblings or with parents can increase competition and if dominant or larger individuals exclude subordinates from resources, it might affect the development of neonates and juveniles. For example, housing neonatal chameleons (*Chamaeleo* spp.) in groups with limited vertical space caused smaller individuals to be forced to the ground, where they reduced their feeding and drinking, to keep their distance from others (Castle 1990). Furthermore, in tree skinks (*Egernia striolata*) housing with a social partner reduced skink growth and subordinate individuals were more likely to suffer tail loss (Riley et al. 2017). Consequently, group housing can be beneficial, but care should be taken in captivity that all individuals have enough space and access to important resources such as food, water, shelter and a thermal and humidity gradient (Burghardt and Layne-Colon 2023).

While the importance of temperature and humidity for captive breeding are well known in reptiles (Burghardt and Layne-Colon 2023; While et al. 2018), far less attention has been given to the social environment, both pre- and post-hatching. Tokay geckos are a large, nocturnal gecko species in which adults perform biparental care towards offspring (Grossmann 2007). Nonetheless, due to their facultative social nature, offspring can be raised isolated from adults. However, it is as of yet unclear what consequences such socially deprived rearing has on the development of offspring or how different levels of competition (number of siblings) might influence growth. Additionally, so far, studies looking at how the incubation environment influences developmental time and post hatching growth are missing in tokay geckos. Therefore, the aim of the current study was to quantify the effects of the incubation environment (humidity and temperature) and the post hatching social environment (group size) on the pre- and post-hatching development of tokay geckos (*Gekko gecko*) during the first six months of life in captivity. Consequently, this study fills an important gap regarding best practice in tokay gecko captive rearing that take their social organisation into account, to ensure best possible welfare in captive bred individuals of this species (Burghardt and Layne-Colon 2023). I hypothesise that both the pre- and post-hatching environment will have an effect on development. I predict that, higher incubation temperature will (1) lead to more males hatching (Gamble 2010; Viets et al. 1994), (2) lead to shorter incubation time (e.g. Elphick and Shine 1998; Noble et al. 2018; Shine et al. 1997b) and (3) might lead to smaller hatchling size due to shorter incubation time (e.g. Van Damme et al. 1992). Furthermore, I predict that rearing in social isolation will have a positive effect on growth due to reduced competition (e.g. Riley et al. 2017).

Methods

Animals

33 captive bred tokay geckos (*Gekko gecko*), 21 females and 12 males, were included in this study. Sex was determined by looking at the presence (for males) or absence (for females) of femoral glands (Grossmann 2007). All animals originated from 16 breeding pairs (see details below) and hatched between May 2022 and March 2023 (first round of breeding) and May and November 2024 (second round of breeding) at the Ethological Station, University of Bern, Bern, Switzerland.

I established 16 breeding pairs from our captive population of 39 adult, captive bred geckos. All adults were purchased from different breeders or bred in house and were between 2 and 8 years old (average = 3.17 years, two females were 8 years old). Adults randomly were paired in January 2022 (first round of breeding) and December 2023 (second round of breeding) and stayed in pairs for a minimum of one year. Females produced their first clutches

(Fig. 1A) in February 2022/ 2023 and continued to produce clutches approximately every 30 days. All eggs and their location were recorded upon discovery of a clutch. Across breeding pairs, 19 individuals hatched from a first clutch, six individuals from a second clutch, one from a fourth clutch, two from a sixth clutch, two from a seventh clutch, and one each from an eighth, tenth and eleventh clutch. This distribution was based on hatching success (some eggs did not develop and were removed by females). All clutches incubated within the home enclosure (see below) in the presence of the parents to ensure natural incubation.

Isolation and group rearing

From around 90 days of incubation, I started checking for hatchlings daily. Offspring started hatching after 78 to 138 days (range) of incubation. After hatching, offspring were allocated to either stay within the home enclosure to be raised with their parents (Figure 1B) or were removed immediately to be raised alone without adults or siblings. Offspring that stayed with the parents either had no siblings or had one sibling or two siblings. Therefore, group sizes ranged from one (isolation, N = 7), three (adult parents only, N = 7), four (adult parents plus one sibling, N = 13) and five (adult parents and two siblings, N = 6). Offspring were raised with parents until they were six months of age and then moved to be housed alone.

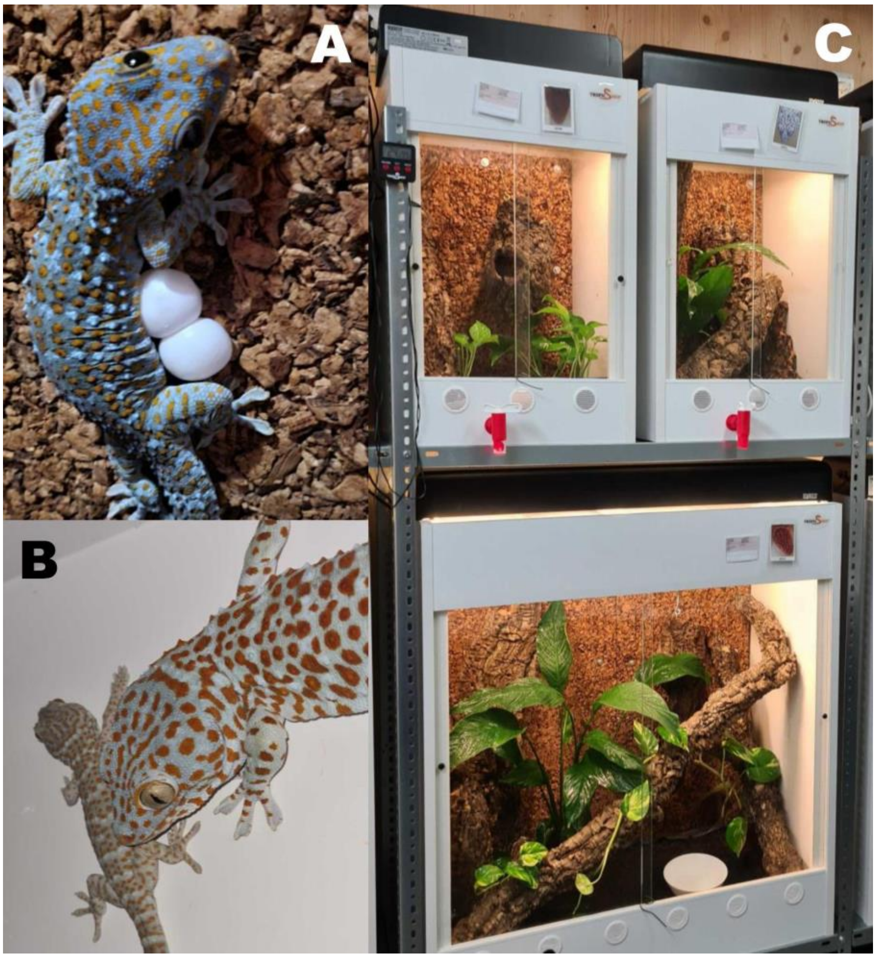

Figure 1. A) Female tokay gecko curling around eggs attached to the cork back wall of the enclosure (shelter removed). B) Male tokay gecko next to one of his offspring (social rearing condition). C) Tank set up showing shelters on the back wall, branches, life plants and a water bowl (picture from 2021). Bottom enclosure for group rearing, top enclosures for single housed females (outside of breeding).

Captive condition

Offspring single housing

Animals were housed in terraria of the size 30 L × 45 B × 45 H cm (only suitable for short term housing in scientific contexts), made of rigid foam plates with a net top and glass front doors. Terraria are fitted with a compressed cork wall fixed to the back, cork branches cut in half hooked on the back (functioning as shelters and nests), cork branches allowing lizards to climb, and life plants as enrichment. Each terrarium has a drainage layer of expanded clay, separated by a mosquito mesh from the soil placed on top (organic tropical forest soil; Dragon BIO-Ground). We spread sphagnum moss and autoclaved red oak leaves on the soil as shelter and food for the isopods that decompose the faecal material of the lizards. Terraria are organized on shelves in three layers. To simulate natural environmental conditions, the room environment is controlled by an automatic system. Animals are exposed to a reversed 12h:12h photo period (i.e. light from 6 pm to 6 am, dark from 6 am to 6 pm). The system imitates sunrise and sunset, which are accompanied by changes in temperature reaching approximately 25 °C during night and 31°C during day. In addition, an UVB light (Exo Terra Reptile UVB 100, 13 W) is provided on top of the terraria during the day. A red light (PHILIPS TL-D 36W/15 RED) invisible to geckos (Loew 1994) is kept on for 24h so as to enable experimenters to work with the lizards. Furthermore, lizards can thermoregulate to their optimal body temperature at any time due to a heat mat (TropicShop) attached to the right outer wall of each enclosure, which locally increases the temperature by 4-5 °C. Humidity is kept at 50 %, but every 12 hours, at 5pm and 4am, 30 seconds of rainfall (with reverse osmotic water) briefly increases humidity to 100%.

Offspring group housing

Terraria for group housing are 90 L × 45 B × 100 H cm in size (only suitable in scientific contexts; Fig. 1C). Except for their size, they are set up exactly the same as the terraria offspring were raised in. Additionally, large enclosures included a larger number of branches and shelters on the back wall, larger plants as well as larger water bowls proportional to the larger enclosure size (Fig. 1C). To prevent small offspring to drown in these water bowls, I added a large stone to ensure easy escape of small individuals.

Husbandry

Offspring were fed five times per week, with 10-15 small to large sized house crickets (*Acheta domesticus*) using scatter feeding. The size of the crickets was adjusted to the changing head size while growing. Adult geckos are fed 3-5 adult house crickets (first round of breeding) or cockroaches (*Nauphoeta cinerea*l; second round of breeding) using 25 cm long forceps in order to control food intake. In order to provide optimal nutrition to our animals (vitamin D and calcium), the insects were fed with high protein dry cat food (various brands, e.g. Purina, Leonardo), cricket mix (reptile planet LDT), and fresh carrots. Fresh water for geckos was supplied ad libitum in water bowls. Moreover, adult geckos were weighted (± 1g) every month and measured (SVL - snout vent length, ±0.5 cm) approximately every three months, to track their body condition.

Measurement of growth and body condition

Every two weeks, offspring were carefully captured by hand and placed in a transparent box (22.5 L × 16.5 B × 7.5 H cm). I then placed a ruler next to the offspring on the outside of the box to take a picture of their ventral side. These pictures were then used to measure SVL using ImageJ 1.53k (Schneider et al., 2012) with the ruler as a reference. Afterwards, offspring were released back into their home enclosure. Hand capture did not lead to tail autotomy in any of the individuals included in this study. At six months of age, offspring were switched to the adult schedule of weighing once a month and measurement of SVL every two months.

Measurement of incubation temperature and humidity

For 32 out of the 33 individuals included in this study, I determined average incubation temperature (day and night pooled), average day and night incubation temperature (separate), average incubation humidity (day and night pooled) as well as average day and night incubation humidity (separate) using 1-Wire® Hygrocgron™ iButtons (DS1923#F5) encased in a waterproof case. iButtons were tied to enclosure furnishings with string to place them directly next to clutches. Temperature and humidity were automatically recorded for a minimum of one week and taken every 15 minutes continuously. In those cases in which measurements were recorded for longer than one week, I extracted averages from a randomly chosen seven consecutive days as our system keeps the humidity and temperature changes stable across time. Finally, I also determined average temperature and humidity in a control location for 10 breeding pairs in breeding round one that was covered by a shelter but where no eggs were deposited. This was done to understand if there are microclimatic conditions (temperature and humidity) that geckos prefer when selecting an egg deposition site. iButtons were again tied to enclosure furnishings to match the height of clutches within the same enclosure.

Ethical statement

The experimental procedure applied in this study was strictly non-invasive and followed the guidelines provided by the Association for the Study of Animal Behaviour/ Animal Behaviour Society for the treatment of animals in behavioural research and Teaching (2023). Experiments were approved by the Suisse Federal Food Safety and Veterinary Office (National No. 33232, Cantonal No. BE144/2020, BE9/2024). Captive conditions were approved by the Suisse Federal Food Safety and Veterinary Office (Laboratory animal husbandry license: No. BE4/11). Three offspring died of natural causes (pathology was inconclusive) during the course of this study. One around 16 weeks and two around six weeks after hatching. During pair formation, we monitored adults closely for 12h to prevent harm. If any aggression occurred, we immediately separated the male and female to avoid injurie. Males were then paired with a different female (N = 17 attempted pairings total) until we established stable pairs that did not show any aggression towards each other. Similarly, after hatching, we monitored hatchlings that stayed with their parents closely and removed one hatchling (G033) due to concerns of insufficient parental care.

Statistical analyses

All statistical analyses were run in R version 4.4.2 (R Core Team 2025). I used Bayesian linear and linear mixed models (LM and LMM) using the package *brms* (Bürkner 2017; 2018; 2021). For all models, I ensured that model Rhat was 1, that the ESS was above 2000 and checked the density plots and correlation plots to check that the models had sampled appropriately. I used a generic weakly informative normal prior and ran 4 chains per model of 5000-7000 iterations each and a thinning interval of 1 (default settings). I used Bayes factors (BF) to evaluate the results by determining Bayes Factors from marginal likelihoods using the package *brms*. Additionally, BF was used to determine if an interaction between fixed effects was significant and if not, it was removed to improve model fit. Bayes factors below 1 indicate no difference, while BF above 1 indicate support for a difference (Schmalz et al. 2023). Data generated during this study and the analysis code are available for download from the Open Science Framework (OSF, link for review purposes: https://osf.io/gdq8y/?view_only=783a534dfbea420b9d2aa9be98913407).

The influence of the incubation environment

I investigated the relationship of average incubation temperature and humidity (response variables in two separate linear models) with offspring sex, incubation time (in days) and SVL at hatching (in cm) (fixed effects). I used the average incubation temperature and humidity because they were highly correlated with both average day (Pearson correlation, temperature: $t = 94.126$, $df = 382$, p-value < 0.001, $r_p = 0.979$; humidity: $t = 346.63$, $df = 382$, p-value < 0.001, $r_p = 0.998$) and night temperature/ humidity (Pearson correlation, temperature: $t = 27.322$, $df = 382$, p-value < 0.001, $r_p = 0.813$; humidity: $t = 339.74$, $df = 382$, p-value < 0.001, $r_p = 0.998$). Incubation time was log transformed as it was not showing a linear relationship with incubation temperature and humidity. In both models, I included a random effect of father identity to account for the fact that multiple offspring were incubated in the same enclosure within and across years (enclosures of males did not change across years).

I also investigated if the average temperature and humidity differed across locations in which geckos deposited their eggs and a control location. I ran two separate linear mixed models, one for the temperature and one for the humidity (response variable) and used location as the only fixed effect. I also included father ID as a random effect to account for measures being taken within the same enclosure (paired data).

The influence of the post-hatching environment

I investigated the relationship of offspring size (SVL, response variable in a linear mixed model) taken across the first six months of life and rearing group size, sex, incubation time and clutch number (fixed effects). To understand if animals showed different growth rates based on rearing group size or sex, I included the interactions of measurement number and rearing group size as well as measurement number and sex as additional fixed effects. Furthermore, I included a random effect of offspring ID nested in father ID to account for repeated measures of SVL within individuals and offspring origin.

To understand the potential long-term effects of the post-hatching environment, I also ran a linear model with the body condition at six months of age (scaled mass index; Peig and Green 2009) (response variable). Offspring that were raised within a family group remained in these family groups for the first six month of their lives. I included rearing group size, SVL at hatching, sex, incubation time and clutch number as fixed effects. Again, I included a random effect of father identity to account for the fact that offspring were raised in their father’s enclosure. I further investigated differences across rearing group sizes using east-squares means (LSM) from the package *emmeans* (Lenth 2025).

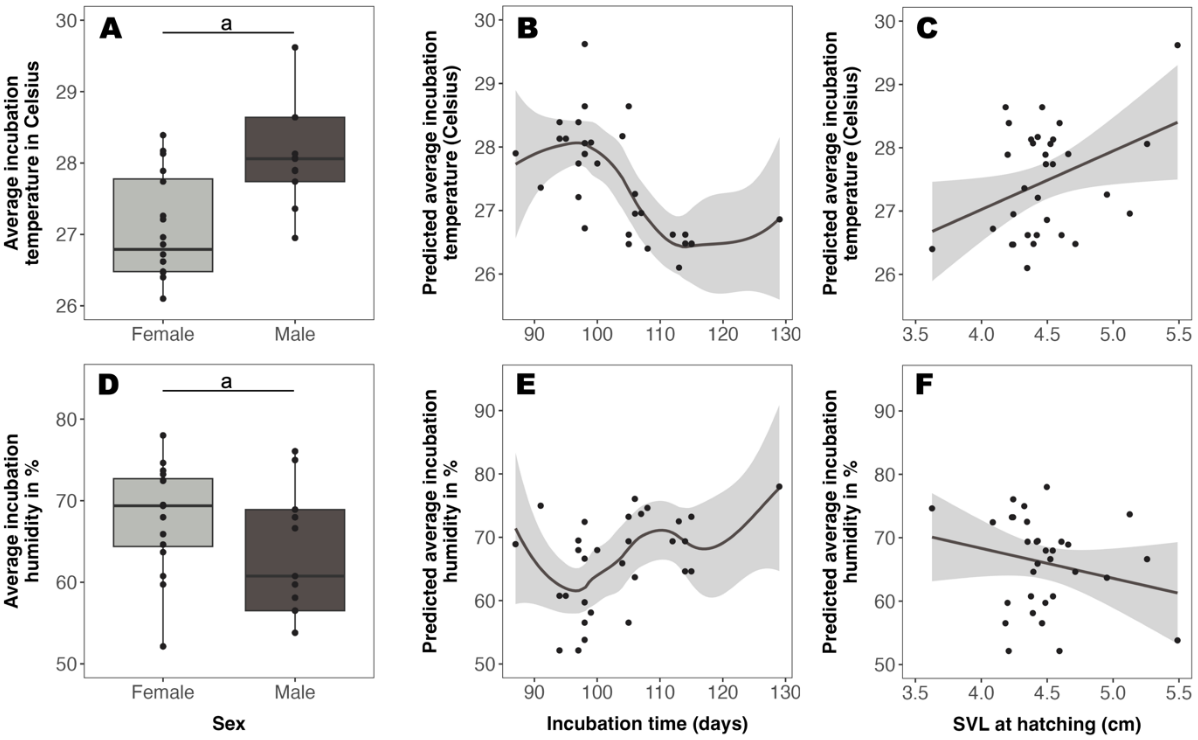

Figure 2. A) Average incubation temperature (in degree Celsius) at which males and females hatch. The bold line within the boxes indicates the median, the upper and lower box edges indicate the upper and lower quartile, and the top and bottom whisker ends indicate the maximum and minimum. B) Predicted relationship between average incubation temperature and incubation time (days). C) Predicted relationship between average incubation temperature and size at hatching (cm). D) Average incubation humidity (%) at which males and females hatch. Dots represent individual data points. E) Predicted relationship between average incubation humidity and incubation time. F) Predicted relationship between average incubation humidity and size at hatching. Across lots, dots represent individual data points and the shaded area around the predicted line represents the 95% confidence interval. a, b – dissimilar letters indicate a difference based on Bayes factors.

Results

The influence of the incubation environment

Average incubation temperature and humidity were negatively correlated (Pearson correlation, t = -23.723, df = 382, p-value < 0.001, r_p = -0.772). I found evidence that more males than females were hatching at higher average incubation temperatures (LM, estimate = 0.437, CI_{low} = 0.326, CI_{up} = 0.549, BF = $2.57*10^{22}$; Fig. 2A). Furthermore, incubation time was also reduced at higher temperatures (LM, estimate = -2.522, CI_{low} = -3.155, CI_{up} = -1.876, BF = $4.01*10^{11}$; Fig. 2B) and hatchlings were larger when incubating at higher average temperatures (LM, estimate = 0.956, CI_{low} = 0.813, CI_{up} = 1.100, BF = $1.82*10^{29}$; Fig. 2C).

I found evidence that more females than males were hatching at higher average incubation humidity (LM, estimate = -2.632, CI_{low} = -3.501, CI_{up} = -1.756, BF = $2.30*10^{18}$; Fig. 2D) and that hatchlings were larger when incubated a higher humidity (LM, estimate = -2.035, CI_{low} = -3.167, CI_{up} = -0.895, BF = 210; Fig. 2F). I found no evidence that average incubation humidity was related to incubation time (LM, estimate = -0.434, CI_{low} = -2.307, CI_{up} = 1.455, BF = 1.062; Fig. 2E).

Finally, I found that average temperature was marginally higher in egg locations compared to control locations (LM, estimate = 0.570, CI_{low} = 0.178, CI_{up} = 0.960, BF = 12) but found no difference in average humidity (LM, estimate = -0.012, CI_{low} = -1.993, CI_{up} = 1.899, BF = 0.994).

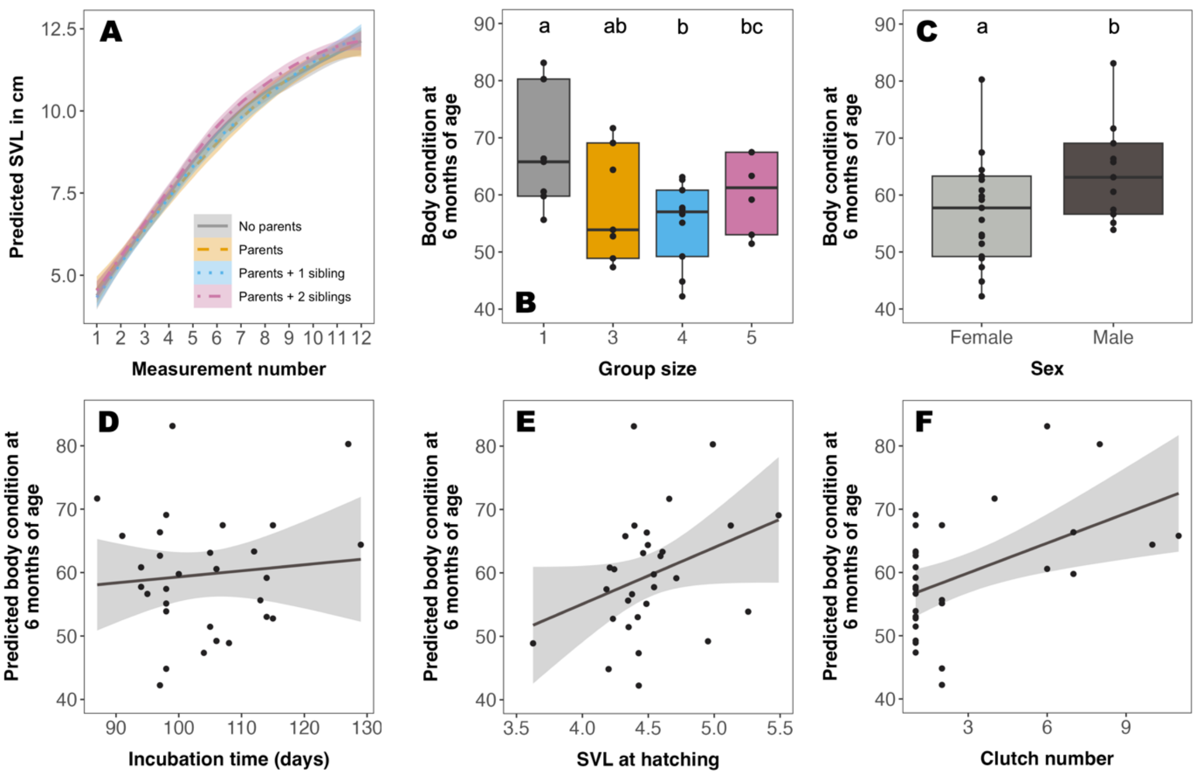

Figure 3. A) Predicted change in SVL across the first six months of life split into the different rearing group sizes. B) Body condition at six months of age split into rearing group sizes. Group sizes ranged from one (isolation, N = 7), three (adult parents only, N = 7), four (adult parents plus one sibling, N = 13) and five (adult parents and two siblings, N = 6). The bold line within the boxes indicates the median, the upper and lower box edges indicate the upper and lower quartiles, and the top and bottom whisker ends indicate the maximum and minimum. C) Body condition of males and females at six months of age. D) Predicted relationship between body condition at six months and incubation time. E) Predicted relationship between body condition at six months and size at hatching. F) Predicted relationship between body condition at six months and clutch number. Across lots, dots represent individual data points and the shaded area around the predicted line represents the 95% confidence interval. a, b, c – dissimilar letters indicate a difference based on Bayes factors (C) or credible interval (B).

The influence of the post-hatching environment

I found evidence that all lizards increased their body size over six months (LMM, estimate = 0.711, $\mathrm{CI}_{\mathrm{low}}$ = 0.692, $\mathrm{CI}_{\mathrm{up}}$ = 0.730, BF = $1.01*10^{211}$; Fig. 3A) but that growth rate did not differ across rearing group sizes (BF = 0.00004, Fig. 3A) or between males and females (BF = 0.054). Therefore, the interactions were removed. I found weak evidence that males were larger than females (LMM, $\text{estimate}_{\text{male}}$ = 0.185, CI_{low} = -0.346, CI_{up} = 0.685, BF = 2.218) but I found no difference across rearing group sizes (LMM, estimate_3 = 0.046, CI_{low} = -0.534, CI_{up} = 0.638; estimate_4 = 0.071, CI_{low} = -0.597, CI_{up} = 0.704; estimate_5 = 0.364, CI_{low} = -0.392, CI_{up} = 1.093; BF = 0.048). Neither incubation time (LMM, estimate = 0.002, CI_{low} = -0.026, CI_{up} = 0.029, BF = 0.014) nor clutch number were associated with SVL (LMM, estimate = 0.043, CI_{low} = -0.046, CI_{up} = 0.131, BF = 0.069).

I also found evidence that the body condition at six months of age differed across rearing group sizes (LMM, estimate_3 = -0.862, CI_{low} = -1.876, CI_{up} = 0.144; estimate_4 = -1.849, CI_{low} = -3.081, CI_{up} = -0.581; estimate_5 = -2.655, CI_{low} = -3.909, CI_{up} = -1.399; BF = 7.821; Fig. 3B). Specifically, individuals that were raised alone had higher body condition compared to individuals that were raised in a family group of at least two siblings and the parents (LSM, Table 1, Fig. 3B). Furthermore, individuals raised with their parents only (group size 3) also had marginally higher body condition than those raised in groups of five (both parents and two siblings, Table 1, Fig. 3B). Moreover, I found that males had higher body condition than females (LM, estimate = 6.755, CI_{low} = 5.731, CI_{up} = 7.748, BF = 2.82×10^{28}; Fig. 3C) and that offspring that incubated longer had higher body condition at six months (LM, estimate = 0.290, CI_{low} = 0.299, CI_{up} = 0.350, BF = 1.56×10^{15}; Fig. 3D). Offspring with a larger hatching size (LM, estimate = 7.829, CI_{low} = 6.646, CI_{up} = 9.013, BF = 1.31×10^{28}; Fig. 3E) and those that hatched from later clutches (LM, estimate = 1.027, CI_{low} = 0.844, CI_{up} = 1.210, BF = 3.34×10^{24}; Fig. 3F) also had better body condition at six months.

Table 1. Least square means difference (estimate) in body condition at six months of age across rearing group sizes. Group sizes ranged from one (isolation, N = 7), three (adult parents only, N = 7), four (adult parents plus one sibling, N = 13) and five (adult parents and

two siblings, N = 6). Contrasts for which the credible interval (CI) does not cross 0 are highlighted in bold.

Group size contrast	Estimate	CI_{low}	CI_{up}
1 versus 3	0.864	-0.124	1.89
1 versus 4	**1.857**	**0.563**	**3.05**
1 versus 5	**2.659**	**1.417**	**3.92**
3 versus 4	0.996	-0.271	2.31
3 versus 5	**1.786**	**0.371**	**3.17**
4 versus 5	0.799	-0.473	2.06

Discussion

In this study, I aimed to gain insights into how incubation temperature and humidity as well as the social environment after hatching influence tokay gecko development. I found that higher temperatures produced more males that were larger and took less time to develop and hatch. Humidity, on the other hand, was not associated with incubation duration but more females that were smaller hatched at higher humidity. However, humidity and temperature were inversely correlated. I also found that geckos selected nest sites with higher temperatures even in captivity, but humidity did not differ between sampling locations. After hatching, hatchling growth did not differ across rearing group size or sexes. Furthermore, I found no relationship between SVL and rearing group size, incubation duration or clutch number, but males were larger than females. Finally, body condition at six months of age, when offspring where still with parents and siblings, was related to rearing group size. Offspring raised alone or in smaller groups had higher body condition than those raised in larger groups. Moreover, body condition was also higher in males, larger hatchlings, in offspring from later clutches, and marginally in hatchlings that incubated longer.

According to my prediction, I found that higher incubation temperatures led to faster development in offspring which is a general trend across all groups of reptiles (Noble et al. 2018). All eggs incubated within the enclosures of the parents, and therefore, did not experience any extreme temperatures (measured range of average temperature = 26.1°C to 29.6°C); despite the small range of only 3.5°C, however, I found a difference in incubation duration of 51 days. None of the offspring hatched with deformities (Burghardt and Layne-Colon 2023; Noble et al. 2018). Therefore, we can assume that the incubation temperatures that individuals experienced were within the appropriate range for optimal development. Also, according to my prediction, I found that more males were hatching at higher incubation temperature, similar to a closely related species, the Schlegel's Japanese Gecko (*Gekko japonicus*; Tokunaga 1985). Again, due to incubation within enclosures, no extreme incubation temperatures were observed, and therefore, I am unable to determine if the sex ratio would have become female biased again at higher temperatures as was found in *G. japonicus* (Tokunaga 1985). Finally, contrary to my prediction hatchlings incubated at higher incubation temperatures were larger possibly because they included more males which were larger than females. According to the literature, in many species no difference in hatching SVL was found at different incubation temperatures (e.g. Elphick and Shine 1998; Shine et al. 1997b; Tiatragul et al. 2017). To disentangle the effect of incubation temperature and sex on hatching size in tokay geckos, future studies should incubate eggs under controlled conditions using temperatures that produce both males and females enabling a direct comparison within each sex. Importantly, I am unaware of a single study investigating the naturally occurring range of incubation temperatures in the wild in this species, important information that could inform investigations into the effects of rising temperatures due to global warming.

Unexpectedly, I found an effect of incubation humidity on hatchling sex. More females were hatching at higher incubation humidity. Similar effects were found in turtles in which higher humidity led to more males hatching (Bell et al. 2025). Both tokay geckos and turtles show temperature dependent sex determination (albeit in opposite directions) and humidity is not often considered as an additional factor aside temperature. However, humidity was negatively correlated with temperature, and it is impossible to tease their effects apart in the current study. Furthermore, I also found an effect of humidity on hatching size in line with a recent meta-analysis that showed a positive relationship between substrate moisture and hatching size with larger hatchlings under moister conditions (Bell et al. 2025). Moreover, contrary to temperature, I found no effect of humidity on incubation duration. Based on my results, tokay geckos could be an interesting model to determine the different effects of temperature and moisture on offspring development in the future.

In line with my last prediction, I found an effect of rearing group size on post-hatching development but only on body condition at six months but not size or growth rate. In my study, offspring were housed in similar, naturalistic enclosures no matter if raised alone or in a family group. Therefore, offspring raised alone only experienced lower social complexity and smaller space compared to offspring raised within a family group. We used scatter feeding of insects during the first six months, and it is possible that offspring raised alone were more successful in finding their prey in smaller enclosures. However, I also found higher body condition in offspring raised in smaller compared to larger family groups, which points more towards differences in competition. Nonetheless, differences in enclosure size together with the lack of competition for food could have given offspring raised in isolation an advantage. In addition to development, in another study, I demonstrated that socially raised individuals were bolder and varied more in their associative learning ability (Szabo and Ringler 2025). Both studies together provide good evidence that sociality, though simpler than in mammals and birds, still has a similar impact on the development of offspring phenotype. In reptiles, it is common to separate neonates after birth or hatching to be raised in simplified environments that facilitate cleaning and welfare checks (Burghardt and Layne-Colon 2023). If such more impoverished environments influence lizard development (physical and/or behavioural), similar to what has been found in birds and mammals (e.g. Meehan and Mench 2002; Schrijver et al. 2002; 2004), is yet unclear. My results also highlight the use of multiple measures to determine the effect of treatment on development as I did find an effect on body condition but not SVL. Generally, more studies are needed to investigate the effects of different housing conditions during development in reptiles (but see Almli and Burghardt 2006; Burghardt and Layne-Colon 2023). Such studies would greatly improve our ability to create optimal captive conditions to produce healthy adult individuals (Burghardt and Layne-Colon 2023). This is not just important in terms of captive welfare but also for reintroduction programs that breed animals in captivity for future release (Crates et al. 2023).

Finally, I was also interested in understanding if tokay geckos select nest sites in captivity despite the lower variability in environmental factors typical for captive environments. As discussed above, it is critical for females to select an appropriate nest location both regarding temperature and humidity to ensure optimal offspring development. In the wild, reptiles show plasticity in their choice of nest site to compensate for unfavorable climatic conditions (e.g. Doody et al. 2006; 2020; Du, et al. 2023) but experiments on nest site selection under more impoverished captive conditions are missing. My results show that geckos select sites with higher temperatures, but humidity seemed unimportant. I also observed that the same nest sites were used by different females across years. Both a temperature and humidity gradient were present in the enclosures, but humidity might have been less pronounced or in line with temperature. Future studies should investigate what conditions geckos prefer when selecting a nest site. Such information would help design better captive environments that meet the animals' needs under multiple contexts.

Conclusion

I found that the incubation environment regarding temperature and humidity and the post-hatching social environment both influenced lizard development. I was, however, not able to also test the effects of clutch size, adjacent developing eggs that might be a source of pre-natal social stimulation, as most eggs incubated in clutches of two. Furthermore, in all cases, eggs incubated in the presence of parents, and it would be interesting to investigate if the absence of parents influences development and hatching success. Thirdly, I have previously shown that females increase basking before egg production (Szabo 2024). This maternal thermal incubation environment embryos experience before oviposition is rarely considered in oviparous reptiles (e.g. McDonald and Schwanz 2018; Schwanz 2016). Future studies could investigate how decreased basking opportunity for females before egg laying influences offspring development which could highlight the need for appropriate basking opportunities for breeding females in captivity. Finally, if tokay gecko juveniles also form dominance hierarchies similar to tree skinks that could explain some of the variation of body condition I observed within rearing treatments is unclear. Importantly, contrary to tree skinks (Riley et al. 2017), I only observed tail loss caused by interactions within the family group in two individuals. Raising multiple offspring without parents, as was done in tree skinks, could shed further light on how sociality might affect juvenile development. Together, my results provide important insights into captive breeding in tokay geckos but many unanswered questions remain.

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