



Grandmaternal age at reproduction affects grandoffspring body condition, reproduction and survival in a wild population of lizards

Josefa Bleu, Sandrine Meylan, Jean Clobert, Manuel Massot

► To cite this version:

Josefa Bleu, Sandrine Meylan, Jean Clobert, Manuel Massot. Grandmaternal age at reproduction affects grandoffspring body condition, reproduction and survival in a wild population of lizards. *Functional Ecology*, 2022, 36 (4), pp.826-836. 10.1111/1365-2435.13996 . hal-03538769

HAL Id: hal-03538769

<https://hal.science/hal-03538769>

Submitted on 7 Feb 2022

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

Grandmaternal age at reproduction affects grandoffspring body condition, reproduction and survival in a wild population of lizards

Josefa Bleu¹, Sandrine Meylan², Jean Clobert³ & Manuel Massot²

¹ Université de Strasbourg, CNRS, Institut Pluridisciplinaire Hubert Curien (UMR 7178), F-67000, Strasbourg, France.

² Sorbonne Université, CNRS, Institut d'Ecologie et des Sciences de l'Environnement de Paris, iEES Paris, F-75005, Paris, France.

³ CNRS, USR 2936, Station d'Écologie Expérimentale du CNRS, route du CNRS, 09200 Moulis, France.

Abstract

1. Age at reproduction can influence the survival and future reproduction of an individual as well as that of their offspring. Remarkably, it has been shown that grandmaternal age at reproduction can also affect the characteristics of grandoffspring in humans and in laboratory or semi-captive animals. However, currently we do not know whether grandmaternal age effects exist in wild populations.
2. We gathered data on female age at reproduction, offspring and grandoffspring characteristics using a 16-year long-term survey of a natural population of the common lizard, *Zootoca vivipara*. The dataset contains 579 grandoffspring from 135 litters.
3. Body size at birth was not correlated with grandmaternal age at reproduction. However, grandoffspring body condition at birth, grandoffspring survival and reproductive performance of granddaughters were dependent on grandmaternal age. These relationships were independent of maternal age.
4. An age-structured model showed that the global effect of grandmaternal age was non linear and was largely driven by its effect on grandoffspring survival. Fitness was higher for granddaughters produced by grandmothers of intermediate ages.

17 5. The study shows that age can shape life-history traits for more than one generation,
18 documenting the importance that grandmaternal age can have in wild populations.

19 **Keywords:** ageing, grandmaternal effect, lagged effect, inter-generational effects, senescence,
20 *Zootoca vivipara*

Introduction

Age at reproduction is known to affect individual reproductive performance, with effects on both litter size and litter success (Clutton-Brock, 1988). First, these results can be explained in terms of the constraints imposed by age. If age is associated with improvements in physiology (*e.g.*, developmental maturation), morphology (*e.g.*, increase in size) or behavior (*e.g.*, knowledge about breeding sites), we expect a positive effect of age in early adulthood (maturation) (Forslund and Pärt, 1995). In contrast, senescence is characterised by physiological deterioration in aged individuals. This process produces higher mortality rates and/or a decrease in reproductive success (Nussey et al., 2013). Second, the age-related performance of an organism can reflect adaptive strategies (Charlesworth & Leon, 1976). Indeed, according to life-history theory, reproductive effort should increase as residual reproductive value declines; this life-history strategy serves to maximise lifetime reproductive success, and we should thus expect a positive effect of age in late adulthood (terminal investment) (Pugesek, 1981; Clutton-Brock, 1984; Roff, 2002).

Interestingly, these constraints and strategies can act at the intra-generational level (*e.g.*, age affects reproductive success of the current generation) or at the inter-generational level with maternal age effects on offspring characteristics (Roff, 2002). Age-related selective pressures can even differ between intra-generational and inter-generational level (Moorad & Nussey, 2016). Even if they are not universal (Moore & Harris, 2003), maternal age effects can be observed on offspring traits at birth (Massot et al., 2011), on offspring survival (Ducatez et al., 2012; Ivimey-Cook & Moorad, 2020) or even on offspring reproductive success (Bouwhuis et al., 2010, 2015; Schroeder et al., 2015). If differences between offspring of the F1 generation (maternal age effects) persist and affect their own offspring in the F2 generation, we may observe grandmaternal (GM) age effects. Inter-generational effects can

create lagged effects in population dynamics and these effects are now recognised as a source of individual heterogeneity that is important to consider to increase our understanding of population dynamics and persistence (Benton et al., 2006). Inter-generational effects can also have evolutionary consequences. They induce time lags in the response of populations to selection, and this may facilitate or hinder adaptive evolution (Kirkpatrick & Lande, 1989; McGlothlin & Galloway, 2014). Importantly, it is crucial to determine whether the effects of age are constant over multiple generations, or cumulative as found in a pioneering study in rotifers (Lansing, 1947), to correctly assess their impact on the population and also to understand whether these effects are indeed adaptive. For instance, adaptive strategies could be selected if age is correlated with environmental variables and thus represents a source of information in itself that could enable to pre-adapt the offspring to the environmental conditions they will encounter. For example, a differential impact of environmental variables (density, resource availability, *etc.*) on survival rates of young and old individuals could correlate age and environmental conditions. For all these possible consequences, it may be important to measure and quantify GM age effects in natural population.

However, few studies have addressed the persistence of age effects over several generations. We are only aware of a few examples in human and some animals studied in laboratory conditions. In human medicine, it has been shown that GM age is a risk factor for trisomy 21 (Papp et al., 1977; Aagesen et al., 1984; Malini & Ramachandra, 2006, but see also, Allen et al., 2009; Kovaleva et al., 2010) but not for breast cancer (de Haan et al., 2010) and that grandpaternal age is positively correlated with telomere size in grandoffspring (Eisenberg et al., 2012, 2019). In non-human animals, the pioneering study in rotifers (Lansing, 1947) showed that the repeated effects of the same maternal age over multiple generations increasingly influenced grandoffspring lifespan, *i.e.*, with evidence of a cumulative effect over multiple generations. A study on aphids (MacKay & Wellington,

1976) showed that GM age influenced grandoffspring reproductive phenotype. More recently, studies on fruit flies have shown that GM age influences grandoffspring viability (Hercus & Hoffmann, 2000; Bloch Qazi et al., 2017), developmental instability (Faurby et al., 2005; Røgilds et al., 2005; Kjarsgaard et al., 2007) and short-term memorisation capacities (Burns & Mery, 2010). GM age effects were also found affecting grandoffspring weight in the rice weevil (Opit & Throne, 2007) and grandoffspring growth and size at maturation in a *Daphnia* (Goos et al., 2019). At the population level, it has also been shown in soil mites that maternal age affects population dynamics for several generations (Benton et al., 2008; Plaistow & Benton, 2009). None of these studies have, however, examined the possibility that such GM age effects occur in natural populations.

In the wild, GM age effects are unexpected for several reasons. First, it has often been assumed that too few individuals survive to advanced ages in the wild to be able to conduct robust statistical analyses on ageing (Kirkwood & Austad, 2000). Second, the high level of environmental variance in nature increases the amount of inter-individual variation (*e.g.*, the variance in resource acquisition is increased), reducing the chance to detect GM age effects. Third, disappearance/appearance of different phenotypes at the population level (*e.g.*, selection) (Forslund & Pärt, 1995; van de Pol et al., 2006) may obscure age effects. Thus, even if age effects are observed under laboratory conditions that enhance the overall survival of individuals, they may not be observed in natural conditions. Although it has been proven possible to measure maternal age effects in the wild (Clutton-Brock, 1988; Nussey et al., 2013), sample sizes to investigate age effects on one additional generation (GM effects) are even more constrained. However, three studies that documented parental effects on the number of grandoffspring recruits showed the feasibility to test GM age effects in wild populations (Bouwhuis et al., 2010, 2015; Schroeder et al., 2015). The constraint on sample size can be circumvented by the long-term monitoring of large populations. Recently, this was

performed by a study on semi-captive Asian elephants in which it was shown a negative GM age effect on grandcalf survival (Reichert et al., 2020). The ultimate challenge for studies that will detect GM age effects in wild populations will be to have enough repeated measurements of the same grandmothers at different ages to discriminate between individual and population level mechanisms (*e.g.*, age-specific selection).

In our study, we tested the effects of grandmaternal age (F0 generation) on the characteristics of grandoffspring at birth (F2 generation), on grandoffspring survival and on the reproduction of granddaughters in a wild population of the common lizard, *Zootoca vivipara* (Figure 1). In this species, maternal age affects reproductive success and offspring characteristics at birth (Massot et al., 2011). In particular, there is a positive effect of female age on female offspring body condition and body size at birth, and this was related both to maturation in young females and terminal investment in old females (Massot et al., 2011). This variation in female offspring quality due to the age of their mother could affect the reproductive outputs of female offspring and lead to GM age effects.

Materials and Methods

Model species and long-term survey

The common lizard (*Zootoca vivipara*, formerly *Lacerta vivipara*) is a small ground-dwelling lizard that is widely distributed across Eurasia. It is a live-bearing species, except in the extreme south of Europe where oviparous populations have been recorded (Surget-Groba et al., 2001). In the study population, the average litter size is five shell-less eggs, and females give birth to fully formed juveniles that emerge from the foetal membrane within one or two hours after laying. There is no parental care, and juveniles are independent from birth. Lizards reproduce once per year, during the summer, and hibernate from late September to April-May. Age-specific survival rates increase until maturity (usually at two years in our population), and

then decrease with age in males and also in females that showed a high reproductive effort during their first reproduction (Massot et al., 2011). The data of our study were obtained from a long-term mark-recapture survey conducted each year from 1989 through 2004 in a natural population at Mont-Lozère (1420 m a.s.l., 44°23'03''N, 3°52'40''E, Cévennes National Park, southeastern France). This population and the long-term survey protocol have been described elsewhere (*e.g.*, Massot & Clobert, 2000; Le Galliard et al., 2010; Massot et al., 2011; Bleu et al., 2013). Briefly, pregnant females are captured each year and kept in the laboratory for three-four weeks until parturition. No sample bias is expected because capture probabilities of adult females do not vary with age (Massot et al., 2011). At birth, offspring are measured (snout-vent length, to the nearest mm), weighed (to the nearest mg), sexed and marked by toe-clipping with no effect on subsequent recapture and survival probabilities (Massot et al., 1992). This protocol allows us to determine the maternal lineages, the age at reproduction, litter size, litter success, offspring body size and body mass at birth, and offspring sex. Lizards that are not born in the laboratory can be aged if they are captured in the field as juveniles or subadults, and this allows us to age many grandmothers. Offspring are recaptured at the end of the reproductive season (in September, *i.e.* before hibernation) and the summer of the next years (in June-July) to estimate their survival. Litters do not always result in live offspring, they can contain stillborns, unfertilised eggs or abortive embryos. We defined litter size as the total litter size, and litter success as the proportion of live offspring in a litter.

Dataset

The full dataset contains 579 grandoffspring from 135 litters, with sampled grandmothers and mothers from two to eight years old (Tables S1 and S2 in Supporting Information). However, in this dataset, some litters were not independent because they were produced by the same grandmother or mother. More precisely, 25 grandmothers and 26 mothers appeared two times in the dataset, five grandmothers and three mothers appeared

three times, four grandmothers and one mother appeared four times and one grandmother appeared five times. The identity of a grandmother or a mother can be repeated for several reasons. Firstly, some litters were produced by different mothers but shared the same grandmother: in eight cases the grandmother had the same age (mothers were from the same litter) and in seven cases the grandmother had different ages (mothers from two different years). The last case is of particular interest because it could allow longitudinal analyses, however it was not very frequent. Secondly, some litters were produced by the same mother at different maternal ages (and thus the same grandmother at the same GM age). This was the most frequent case, with 26 cases where the mother was captured two different years, three cases three different years and one case four different years.

Statistical analyses

We tested the effects of grandmaternal (GM) age on grandoffspring characteristics at birth and on the reproduction of granddaughters with mixed and linear models using R 4.1.1 statistical software (R Core Team, 2021). The significance of the fixed effects was tested using F statistics for linear mixed models and a chi-square test for logistic regression (Bolker et al., 2009). We checked the homoscedasticity and the normality of the residuals of the linear mixed models. The collinearity of the explanatory variables was also checked using variance inflation factors.

Firstly, we analysed the effects of GM age on grandoffspring body size at birth (snout-vent length) and on grandoffspring body condition at birth. We defined body condition as the residuals of a linear regression of body mass against body size. The regression was performed for each sex separately because of sexual size dimorphism in juvenile common lizards (Le Galliard et al., 2006). We used the ‘*lmer*’ function from the packages ‘*lme4*’ (version 1.1-27) (Bates et al., 2015) and ‘*lmerTest*’ (version 3.1-3) (Kuznetsova et al., 2017) to compute linear mixed models with the Satterthwaite's method to compute degrees of freedom and F-statistics.

The explanatory variables of the full models were: GM age, maternal age (M age), offspring sex and the first-order interactions. Because age effects are often curvilinear (*e.g.*, Massot et al., 2011), the quadratic terms of the GM age and M age were also included in the model. The random effects were the year of maternal reproduction and maternal identity (offspring from the same litter are not independent and some mothers may appear different years in the dataset). The identity of the grandmother was not included as a random effect because the variance explained by this effect was close to 0. We believe that it might not be relevant to include this effect as we did not have enough repetitions of grandoffspring from the same grandmother but different mothers to estimate this properly. We also did not have individual estimates of longevity of mothers and grandmothers to include as covariates to control for selective disappearance related to maternal and grandmaternal ages (*e.g.*, Hayward et al., 2013).

Secondly, we analysed reproduction of granddaughters at the age of two years old, *i.e.* at their first reproduction (Massot et al. 2011). We analysed granddaughter litter size and granddaughter litter success with logistic regression (generalized linear models). For litter size, we used a Conway-Maxwell-Poisson regression to take into account underdispersion of these count data (dispersion parameter = 0.26) (Sellers & Premeaux, 2020). More precisely, we used the ‘*glm.cmp*’ function from the package ‘*mpcmp*’ (version 0.3.6). For litter success, we used a logistic regression with quasi-binomial error to take into account overdispersion of these proportion data (dispersion parameter = 1.66). We used the ‘*glm*’ function (base package) and “*Anova*” from the car package (version 3.0-11) (Fox & Weisberg, 2019) to compute type 3 anova. Because of the small sample sizes, we tested only the interaction between GM age and M age and we did not add random effects (the identity of only one mother and two grandmothers were repeated in the dataset). The explanatory variables of the full model were: GM age (linear and quadratic terms), M age (linear and quadratic terms), and

granddaughter body size. We included granddaughter body size because reproduction depends on body size in female common lizards (Massot et al., 1992, 2011).

Survival analyses

Grandoffspring were recaptured at the end of the summer before their first hibernation and each summer of the next years. The resulting capture-mark-recapture data allowed us to estimate the survival probabilities. Survival estimates were obtained while taking into account the capture probabilities using a capture-mark-recapture method based on the open population model of Cormack-Jolly-Seber (Clobert et al., 1987). This model produces apparent survival estimates resulting from mortality and emigration. Because we have shown that emigration is rare in our study population, our estimates may be assimilated to true survival rates (Massot et al., 1992). We used the program E-surge version 2.2.3 to fit models (Choquet et al., 2009b). We tested whether survival and capture probabilities were dependent on GM age and M age (linear and quadratic terms), and the interaction between the two. The full model of survival probabilities also included as factors the season (autumn/summer), the year and the sex in interaction with age classes (variation in survival between age classes are reported in Massot et al., 2011). Concerning capture probabilities, the full model included the season, year and sex factors in addition to the variables of GM age and M age (linear and quadratic terms, and their interaction). We first selected a model for capture probabilities (while keeping the full model for survival probabilities) and then a model for survival probabilities. The model selection was based on the quasi-likelihood Akaike information criterion (QAICc), this criterion accounts for the overdispersion of data that can be induced by missing factors (Burnham & Anderson, 2002). The goodness of fit of the Cormack-Jolly-Seber model to the data was checked with the program U-CARE version 2.3.4 (Choquet et al., 2009a).

Age-structured matrix model

In order to approximate the GM age effect on granddaughter fitness, we implemented an age-structured matrix model with the ULM software version 6.0 (Legendre & Clobert, 1995). We modelled the life cycle of the common lizard from a 9-age class Leslie matrix, and we calculated the asymptotic multiplication rate as the dominant eigenvalue of the matrix. The asymptotic multiplication rate was used as a proxy of fitness. As usual for this kind of modelling, our model was based solely on females. This approach assumes that male abundance is always high enough to ensure reproduction of most females (Bessa-Gomes et al., 2010). This assumption is relevant to our studied population where all sexually mature females (females of three years of age and older, and two years old females with a body size larger than a threshold) reproduce (Massot et al., 2011). We fixed the female age at maturity at two years as usually observed (Massot et al., 2011). The proportion of females at birth was fixed at 50% (Le Galliard et al., 2005) to adjust fecundities to the numbers of females produced. The values of age-specific fecundity of granddaughters and survival for the first winter were the estimates from this study (Figure 1). The ULM software allows to compute the age-specific multiplication rates and to measure the multiplication rates sensitivity to change in the different vital parameters tested (Legendre & Clobert, 1995; Caswell, 2001). The ULM software can be downloaded from <http://www.biologie.ens.fr/~legendre/ulm/ulm.html>.

Results

Body size and body condition at birth of grandoffspring

Table 1 shows the results of the analysis on body size. Body size at birth of F2 individuals was dependent on maternal age (F1 generation) and grandoffspring sex. F2 females were bigger than F2 males and the effect of maternal age was quadratic with a

maximum for mothers of five years old (Figure S1). However, there was no significant effect of GM age (F0 generation).

Body condition at birth of F2 individuals was dependent on maternal age (F1 generation) and GM age (F0 generation) (Table 2). Body condition at birth was dependent on maternal age in interaction with sex. F2 males had lower body condition than F2 females from older mothers (Figure S1). Body condition of F2 individuals was also significantly related to GM age in interaction with sex (Table 2). Body condition decreased with GM age for F2 males (estimate \pm s.e. = -1.27 ± 1.07) but not for F2 females (estimate = 0.60 ± 1.10). As a consequence, F2 males had lower body condition than F2 females from older grandmothers (Figure 2).

Survival of grandoffspring

We studied M age (F1) and GM age (F0) effects on survival probabilities of grandoffspring (F2 individuals). Figure 3 shows survival probabilities of grandoffspring during their first hibernation, and the range of 95% confidence interval of survival probabilities for all the combinations of grandmaternal and maternal ages. Survival probabilities of grandoffspring had large confidence intervals mainly in the combinations of the oldest grandmothers and mothers (right panels of Figure 3). Grandoffspring survival was not confidently estimated in these combinations of the oldest ages as we could expect from their small sample sizes (Table S2). Figure 3 (left panels) reports only survival probabilities of grandoffspring the most confidently estimated. Overall, we found an effect of both M age (linear effect) and GM age (quadratic effect) (log-likelihood ratio tests: $X^2_1 = 6.1$ $P = 0.013$ for M age effect, $X^2_1 = 8.1$ $P = 0.005$ for GM age quadratic effect; detailed analysis in Table S3). The maternal and grandmaternal effects were additive. Male and female offspring of the oldest mothers showed the highest survival rates (left panels of Figure 3). For the quadratic

GM age effect, the survival rates of male and female grandoffspring were the highest for the three years old grandmothers (left panels of Figure 3).

Reproduction of granddaughters

Twenty-two pregnant granddaughters were recaptured at the age of two years old, the age at which females may first reproduce in this population (Massot et al., 2011). Litter size and litter success of granddaughters (F2 females) were dependent on GM age (F0 females) (Tables 3 and 4). The reproductive performances of granddaughters increased with GM age (Figure 4). Granddaughter litter size was also positively correlated with granddaughter body size (Table 3). Granddaughter litter size and success were not significantly related to maternal age (Tables 3 and 4).

Age-structured population model

We calculated the net effect of GM age effects by estimating the multiplication rate (λ) of granddaughters produced by GM of different ages with an aged-structured matrix model. This measure is an approximation of the fitness of granddaughters since it takes into account both the GM age effects on survival and fecundity. GM age has a non linear effect on λ , with maximum values for three and four years old GM (Figure 5). λ is clearly smaller for the granddaughters produced by grandmothers older than four years old (Figure 5). The sensitivity was 0.71 for juvenile survival, 0.05 for fecundity at age 2, 0.03 for fecundity at age 3, and less than 0.02 for fecundity at older ages. This indicates that most of this pattern is driven by the GM effects on grandoffspring survival.

Discussion

A recent review of animal populations has illustrated that offspring quality often depends on maternal traits (Moore et al., 2019). The literature on maternal effects expanded greatly over the last three decades and showed that these effects are common and affect a

diversity of traits. Specifically, a series of previous studies in the common lizard found that this live-bearing species exhibits multiple maternal effects. For example, offspring body size and body condition were dependent on maternal age (Massot et al., 2011), offspring growth and survival were influenced by temperature during gestation (Lorenzon et al., 2001), offspring dispersal responded to food availability during gestation (Massot et al., 2008), and litter size was related to rainfall during vitellogenesis (Bleu et al., 2013). The commonness of maternal effects in the common lizard might explain why we found grandmaternal age effects on four out of the five parameters tested, with GM age effects on body condition at birth, survival and two reproductive parameters. Interestingly, these grandmaternal age effects were independent of maternal age effects. GM age and M age had only additive effects on grandoffspring body condition and survival. We found only a GM age effect on granddaughter reproduction. We found no interaction between GM age and M age effects for all the tested parameters (*i.e.*, no M age x GM age interaction in Tables 1 to 4, and Table S3). Therefore, we did not detect a cumulative effect of maternal age effects over multiple generations as in the pioneering study of Lansing (1947).

Our study illustrates that the effects of age at reproduction can persist for up to two generations in a wild population, with age effects on grandoffspring body condition, survival and reproduction. Age effects may be explained by individual consequences of ageing (maturation, senescence and terminal investment) or by the age-specific disappearance/appearance of different phenotypes at the population level (*e.g.*, selection) (Forslund & Pärt, 1995; van de Pol et al., 2006). In this study, we did not have enough data to perform longitudinal analyses with repeated measurements of the same grandmothers at different ages. We also did not have the possibility to use individual estimates of longevity of mothers and grandmothers as covariates to control for selective disappearance related to maternal and grandmaternal ages (*e.g.*, Hayward et al., 2013). For this reason, we analysed

only the patterns at the population level and we cannot formally distinguish between individual and population effects. A previous study in the common lizard supports the hypothesis of responses at the individual level for at least a part of the grandmaternal age effects we observed. This study performed analyses on longitudinal data with repeated measurements of the same mothers at different ages (Massot et al., 2011) and found relationships between maternal age and offspring traits at birth (body size and body condition). However, we cannot know if these individual responses at the first generation span the next generation. We also cannot rule out the possibility that these individual responses were modified or replaced by a differential mortality among offspring of the first generation (hypothesis of selective disappearance at the population level).

Yet, whatever the levels of action, this study shows that variation of the age at reproduction had effects lasting for up to two generations. Although it would be interesting to understand the mechanisms, finding an overall GM age effect is a first and necessary step. This shows that GM age effects are a reality in wild populations that can affect population dynamics, and that they are not just a human specificity or a laboratory curiosity. This is consistent with a study on semi-captive Asian elephants (Reichert et al., 2020) in which it has been shown that grandoffspring of old grandmothers had reduced survival (Reichert et al., 2020). Our study found also that male and female grandoffspring of old grandmothers had reduced survival. Maternal effects were also observed in elephants. Offspring of older mothers had reduced survival and higher reproductive success. In addition, it was suggested a quadratic effect of maternal age on offspring body condition. Because offspring of older mothers had reduced survival, the authors discussed the hypothesis that the differential mortality between F0 and F1 generations led to the GM age effect at the F2 generation (Reichert et al., 2020). As in our study, it appeared challenging to discriminate between individual and population level mechanisms.

In our study, we analysed long-term GM effects on grandoffspring survival and reproduction. The effect of GM age on survival was globally negative with a decrease of grandoffspring survival when grandmothers were older than four years old (Figure 3). For the fecundity of granddaughters, the effect of GM age was positive. Litter size and litter success of granddaughters increased with increasing GM age (Figure 4). To understand the combined effects of GM age on survival and fecundity, we compared the multiplication rates of granddaughters produced by grandmothers of different ages. The overall pattern shows a non linear trend with a maximum of fitness for the granddaughters of grandmothers of three and four years old (Figure 5). Even if it is difficult to conclude on the adaptive value of the GM effects detected since we do not know if they are at the population or individual levels, these GM age effects seem concordant with individual responses related to maturation in young grandmothers and senescence in old grandmothers.

In conclusion, we found evidence that the age of grandmothers at reproduction (F0 generation) has effects on grandoffspring (F2 generation) in a wild population. These effects that persist for up to two generations induce time lags in the response of populations to selective pressures, and may facilitate or hinder adaptive evolution (Kirkpatrick & Lande, 1989; McGlothlin & Galloway, 2014). Our study provides two useful avenues for future research. First, it will be important to understand the mechanisms underlying the grandmaternal age effects, especially determining if they are individual or/and population responses. Second, it would be interesting to test whether the age of the grandfathers at reproduction may also have effects in wild populations, as found in humans (Eisenberg et al., 2012, 2019).

Acknowledgments: We thank all students and colleagues who helped collecting data over the years, the Parc National des Cévennes and the Office National des Forêts for field work facilities. This work was supported by the Agence Nationale de la Recherche (grant 07-

BLAN-0217 to M.M. and J.C.).

Conflict of interests: The authors declare no conflict of interest.

Authors' contributions: All authors conceived the ideas. MM collected the data. JB analysed the data and led the writing of the manuscript. All authors contributed critically to the drafts and gave final approval for publication.

Data availability statement: Data deposited in the Dryad Digital Repository: <https://doi.org/10.5061/dryad.9ghx3ffjv>, (Bleu et al., 2021).

References

- Aagesen, L., Grinsted, J., & Mikkelsen, M. (1984). Advanced grandmaternal age on the mother's side - a risk of giving rise to trisomy 21. *Annals of Human Genetics*, 48(4), 297–301. <https://doi.org/10.1111/j.1469-1809.1984.tb00843.x>
- Allen, E. G., Freeman, S. B., Druschel, C., Hobbs, C. A., O'Leary, L. A., Romitti, P. A., ... Sherman, S. L. (2009). Maternal age and risk for trisomy 21 assessed by the origin of chromosome nondisjunction: a report from the Atlanta and national Down syndrome projects. *Human Genetics*, 125(1), 41–52. <https://doi.org/10.1007/s00439-008-0603-8>
- Bates, D., Mächler, M., Bolker, B. M., & Walker, S. C. (2015). Fitting linear mixed-effects models using lme4. *Journal of Statistical Software* 67(1), 1–48. <https://doi.org/10.18637/jss.v067.i01>
- Benton, T. G., Plaistow, S. J., & Coulson, T. N. (2006). Complex population dynamics and complex causation: devils, details and demography. *Proceedings of the Royal Society B: Biological Sciences*, 273(1591), 1173–1181. <https://doi.org/10.1098/rspb.2006.3495>
- Benton, T. G., St Clair, J. J. H., & Plaistow, S. J. (2008). Maternal effects mediated by maternal age: from life histories to population dynamics. *Journal of Animal Ecology*, 77(5), 1038–1046. <https://doi.org/10.1111/j.1365-2656.2008.01434.x>
- Bessa-Gomes, C., Legendre, S., & Clobert, J. (2010). Discrete two-sex models of population dynamics: on modelling the mating function. *Acta Oecologica*, 36(5), 439–445. <https://doi.org/10.1016/j.actao.2010.02.010>

398 Bleu, J., Le Galliard, J.-F., Fitze, P., Meylan, S., Clobert, J., & Massot, M. (2013). Reproductive allocation
399 strategies: a long-term study on proximate factors and temporal adjustments in a viviparous lizard.
400 *Oecologia*, 171, 141–151. <https://doi.org/10.1007/s00442-012-2401-1>

401 Bloch Qazi, M. C., Miller, P. B., Poeschel, P. M., Phan, M. H., Thayer, J. L., & Medrano, C. L. (2017).
402 Transgenerational effects of maternal and grandmaternal age on offspring viability and performance in
403 *Drosophila melanogaster*. *Journal of Insect Physiology*, 100, 43–52.
404 <https://doi.org/10.1016/j.jinsphys.2017.05.007>

405 Bolker, B. M., Brooks, M. E., Clark, C. J., Geange, S. W., Poulsen, J. R., Stevens, M. H. H., & White, J.-S. S.
406 (2009). Generalized linear mixed models: a practical guide for ecology and evolution. *Trends in*
407 *Ecology & Evolution*, 24(3), 127–135. <https://doi.org/10.1016/j.tree.2008.10.008>

408 Bouwhuis, S., Charmantier, A., Verhulst, S., & Sheldon, B. C. (2010). Trans-generational effects on ageing in a
409 wild bird population. *Journal of Evolutionary Biology*, 23(3), 636–642. [https://doi.org/10.1111/j.1420-](https://doi.org/10.1111/j.1420-9101.2009.01929.x)
410 [9101.2009.01929.x](https://doi.org/10.1111/j.1420-9101.2009.01929.x)

411 Bouwhuis, S., Vedder, O., & Becker, P. H. (2015). Sex-specific pathways of parental age effects on offspring
412 lifetime reproductive success in a long-lived seabird. *Evolution*, 69(7), 1760–1771.
413 <https://doi.org/10.1111/evo.12692>

414 Burnham, K. P., & Anderson, D. R. (2002). *Model Selection and Multimodel Inference: a practical information-*
415 *theoretic approach* (2nd ed.). New-York, NY: Springer US.

416 Burns, J. G., & Mery, F. (2010). Transgenerational memory effect of ageing in *Drosophila*. *Journal of*
417 *Evolutionary Biology*, 23(4), 678–686. <https://doi.org/10.1111/j.1420-9101.2010.01932.x>

418 Caswell, H. (2001). *Matrix population models: construction, analysis, and interpretation*. Sunderland,
419 Massachusetts: Sinauer Associates.

420 Charlesworth, B., & Leon J. A. (1976). The relation of reproductive effort to age. *The American Naturalist*,
421 110(973), 449-459. <https://doi.org/10.1086/283079>

422 Choquet, R., Lebreton, J.-D., Gimenez, O., Reboulet, A.-M., & Pradel, R. (2009a). U-CARE: Utilities for
423 performing goodness of fit tests and manipulating Capture-REcapture data. *Ecography*, 32(6), 1071–
424 1074. <https://doi.org/10.1111/j.1600-0587.2009.05968.x>

425 Choquet, R., Rouan, L., & Pradel, R. (2009b). Program E-Surge: a software application for fitting multievent
426 models. In D. L. Thomson, E. G. Cooch, & M. J. Conroy (Eds.), *Modeling Demographic Processes In*
427 *Marked Populations* (pp. 845–865). New-York, NY: Springer US.

428 Clobert, J., Lebreton, J.-D., & Allainé, D. (1987). A general approach to survival rate estimation by recaptures or
 429 resightings of marked birds. *Ardea*, 75(1), 133–142.

430 Clutton-Brock, T. H. (1984). Reproductive effort and terminal investment in iteroparous animals. *The American*
 431 *Naturalist*, 123(2), 212–229. <https://doi.org/10.1086/284198>

432 Clutton-Brock, T. H. (1988). *Reproductive success: studies of individual variation in contrasting breeding*
 433 *systems*. University of Chicago Press.

434 de Haan, M. C., Michels, K. B., Peeters, P. H. M., van Noord, P. a. H., Hennekam, F. a. M., & van der Schouw,
 435 Y. T. (2010). Age of mother and grandmother in relation to a subject's breast cancer risk. *British*
 436 *Journal of Cancer*, 102(9), 1400–1404. <https://doi.org/10.1038/sj.bjc.6605639>

437 Ducatez, S., Baguette, M., Stevens, V. M., Legrand, D., & Freville, H. (2012). Complex interactions between
 438 paternal and maternal effects: parental experience and age at reproduction affect fecundity and offspring
 439 performance in a butterfly. *Evolution*, 66(11), 3558–3569. [https://doi.org/10.1111/j.1558-](https://doi.org/10.1111/j.1558-5646.2012.01704.x)
 440 [5646.2012.01704.x](https://doi.org/10.1111/j.1558-5646.2012.01704.x)

441 Eisenberg, D. T. A., Hayes, M. G., & Kuzawa, C. W. (2012). Delayed paternal age of reproduction in humans is
 442 associated with longer telomeres across two generations of descendants. *Proceedings of the National*
 443 *Academy of Sciences*, 109(26), 10251–10256. <https://doi.org/10.1073/pnas.1202092109>

444 Eisenberg, D. T. A., Lee, N. R., Rej, P. H., Hayes, M. G., & Kuzawa, C. W. (2019). Older paternal ages and
 445 grandpaternal ages at conception predict longer telomeres in human descendants. *Proceedings of the*
 446 *Royal Society B: Biological Sciences*, 286(1903), 20190800. <https://doi.org/10.1098/rspb.2019.0800>

447 Faurby, S., Kjærsgaard, A., Pertoldi, C., & Loeschcke, V. (2005). The effect of maternal and grandmaternal age
 448 in benign and high temperature environments. *Experimental Gerontology*, 40(12), 988–996.
 449 <https://doi.org/10.1016/j.exger.2005.08.007>

450 Forslund, P., & Pärt, T. (1995). Age and reproduction in birds - hypotheses and tests. *Trends in Ecology &*
 451 *Evolution*, 10(9), 374–378. [https://doi.org/10.1016/S0169-5347\(00\)89141-7](https://doi.org/10.1016/S0169-5347(00)89141-7)

452 Fox, J., & Weisberg, S. (2019). *An R Companion to Applied Regression* (3rd ed.). SAGE Publications.

453 Goos, J. M., Swain, C. J., Munch, S. B., & Walsh, M. R. (2019). Maternal diet and age alter direct and indirect
 454 relationships between life-history traits across multiple generations. *Functional Ecology*, 33(3), 491–
 455 502. <https://doi.org/10.1111/1365-2435.13258>

456 Hayward, A. D., Wilson A. J., Pilkington, J. G., Clutton-Brock, T. H., Pemberton, J. M., & Kruuk, L. E. B.
 457 (2013). Reproductive senescence in female Soay sheep: variation across traits and contributions of

individual ageing and selective disappearance. *Functional Ecology*, 27 (1), 184–195.

<https://doi.org/10.1111/1365-2435.12029>

Hercus, M. J., & Hoffmann, A. A. (2000). Maternal and grandmaternal age influence offspring fitness in *Drosophila*. *Proceedings of the Royal Society of London. Series B: Biological Sciences*, 267(1457), 2105–2110. <https://doi.org/10.1098/rspb.2000.1256>

Ivimey-Cook, E., & Moorad, J. (2020). The diversity of maternal-age effects upon pre-adult survival across animal species. *Proceedings of the Royal Society B: Biological Sciences* 287 (1932): 20200972. <https://doi.org/10.1098/rspb.2020.0972>

Kirkpatrick, M., & Lande, R. (1989). The evolution of maternal characters. *Evolution*, 43(3), 485–503. <https://doi.org/10.2307/2409054>

Kirkwood, T. B. L., & Austad, S. N. (2000). Why do we age? *Nature*, 408(6809), 233–238. <https://doi.org/10.1038/35041682>

Kjarsgaard, A., Faurby, S., Holmandersen, D. H., Pertoldi, C., David, J. R., & Loeschcke, V. (2007). Effects of temperature and maternal and grandmaternal age on wing shape in parthenogenetic *Drosophila mercatorum*. *Journal of Thermal Biology*, 32(2), 59–65. <https://doi.org/10.1016/j.jtherbio.2006.10.001>

Kovaleva, N. V., Tahmasebi-Hesari, M., & Verlinskaya, D. K. (2010). Grandmaternal ages at birth of parents of children with Down syndrome in St. Petersburg. *Cytology and Genetics*, 44(5), 300–305. <https://doi.org/10.3103/S0095452710050075>

Kuznetsova, A., Brockhoff, P. B., & Christensen, R. H. B. (2017). lmerTest Package: Tests in Linear Mixed Effects Models. *Journal of Statistical Software*, 82(13), 1–26. <https://doi.org/10.18637/jss.v082.i13>

Lansing, A. I. (1947). A transmissible, cumulative, and reversible factor in aging. *Journal of Gerontology*, 2(3), 228–239. <https://doi.org/10.1093/geronj/2.3.228>

Le Galliard, J.-F., Fitze, P. S., Cote, J., Massot, M., & Clobert, J. (2005). Female common lizards (*Lacerta vivipara*) do not adjust their sex-biased investment in relation to the adult sex ratio. *Journal of Evolutionary Biology*, 18(6), 1455–1463. <https://doi.org/10.1111/j.1420-9101.2005.00950.x>

Le Galliard, J.-F., Marquis, O., & Massot, M. (2010). Cohort variation, climate effects and population dynamics in a short-lived lizard. *Journal of Animal Ecology*, 79(6), 1296–1307. <https://doi.org/10.1111/j.1365-2656.2010.01732.x>

486 Le Galliard, J.-F., Massot, M., Landys, M. M., Meylan, S., & Clobert, J. (2006). Ontogenic sources of variation
 487 in sexual size dimorphism in a viviparous lizard. *Journal of Evolutionary Biology*, 19(3), 690-704.
 488 <https://doi.org/10.1111/j.1420-9101.2006.01094.x>

489 Legendre, S., & Clobert, J. (1995). ULM, a software for conservation and evolutionary biologists. *Journal of*
 490 *Applied Statistics*, 22(5-6), 817–834. <https://doi.org/10.1080/02664769524649>

491 Lorenzon, P., Clobert, J., & Massot, M. (2001). The contribution of phenotypic plasticity to adaptation in
 492 *Lacerta vivipara*. *Evolution*, 55(2), 392–404. [https://doi.org/10.1554/0014-](https://doi.org/10.1554/0014-3820(2001)055[0392:TCOPPT]2.0.CO;2)
 493 [3820\(2001\)055\[0392:TCOPPT\]2.0.CO;2](https://doi.org/10.1554/0014-3820(2001)055[0392:TCOPPT]2.0.CO;2)

494 MacKay, P. A., & Wellington, W. G. (1976). Maternal age as a source of variation in the ability of an aphid to
 495 produce dispersing forms. *Researches on Population Ecology*, 18(1), 195–209.
 496 <https://doi.org/10.1007/BF02754093>

497 Malini, S. S., & Ramachandra, N. B. (2006). Influence of advanced age of maternal grandmothers on Down
 498 syndrome. *BMC Medical Genetics*, 7(1), 4. <https://doi.org/10.1186/1471-2350-7-4>

499 Massot, M., & Clobert, J. (2000). Processes at the origin of similarities in dispersal behaviour among siblings.
 500 *Journal of Evolutionary Biology*, 13(4), 707–719. <https://doi.org/10.1046/j.1420-9101.2000.00202.x>

501 Massot, M., Clobert, J., & Ferrière, R. (2008). Climate warming, dispersal inhibition and extinction risk. *Global*
 502 *Change Biology*, 14(3), 461–469. <https://doi.org/10.1111/j.1365-2486.2007.01514.x>

503 Massot, M., Clobert, J., Montes-Poloni, L., Haussy, C., Cubo, J., & Meylan, S. (2011). An integrative study of
 504 ageing in a wild population of common lizards. *Functional Ecology*, 25(4), 848–858.
 505 <https://doi.org/10.1111/j.1365-2435.2011.01837.x>

506 Massot, M., Clobert, J., Pilorge, T., Lecomte, J., & Barbault, R. (1992). Density dependence in the common
 507 lizard: demographic consequences of a density manipulation. *Ecology*, 73(5), 1742–1756.
 508 <https://doi.org/10.2307/1940026>

509 McGlothlin, J. W., & Galloway, L. F. (2014). The contribution of maternal effects to selection response: an
 510 empirical test of competing models. *Evolution*, 68(2), 549–558. <https://doi.org/10.1111/evo.12235>

511 Moorad, J. A., & Nussey D. H. (2016). Evolution of maternal effect senescence. *Proceedings of the National*
 512 *Academy of Sciences*, 113 (2), 362-367. <https://doi.org/10.1073/pnas.1520494113>

513 Moore, M. P., Whiteman, H. H., & Martin, R. A. (2019). A mother's legacy: the strength of maternal effects in
 514 animal populations. *Ecology Letters*, 22(10), 1620–1628. <https://doi.org/10.1111/ele.13351>

515 Moore, P. J., & Harris, W. E. (2003). Is a decline in offspring quality a necessary consequence of maternal age?
 516 *Proceedings of the Royal Society of London B: Biological Sciences*, 270(S2), S192–S194.
 517 <https://doi.org/10.1098/rsbl.2003.0051>

518 Nussey, D. H., Froy, H., Lemaître, J.-F., Gaillard, J.-M., & Austad, S. N. (2013). Senescence in natural
 519 populations of animals: widespread evidence and its implications for bio-gerontology. *Ageing Research*
 520 *Reviews*, 12(1), 214–225. <https://doi.org/10.1016/j.arr.2012.07.004>

521 Opit, G. P., & Throne, J. E. (2007). Influence of maternal age on the fitness of progeny in the rice weevil,
 522 *Sitophilus oryzae* (Coleoptera: Curculionidae). *Environmental Entomology*, 36(1), 83–89.
 523 [https://doi.org/10.1603/0046-225X\(2007\)36\[83:IOMAOT\]2.0.CO;2](https://doi.org/10.1603/0046-225X(2007)36[83:IOMAOT]2.0.CO;2)

524 Papp, Z., Varadi, E., & Szabo, Z. (1977). Grandmaternal age at birth of parents of children with trisomy 21.
 525 *Human Genetics*, 39(2), 221–224. <https://doi.org/10.1007/BF00287015>

526 Plaistow, S. J., & Benton, T. G. (2009). The influence of context-dependent maternal effects on population
 527 dynamics: an experimental test. *Philosophical Transactions of the Royal Society B: Biological Sciences*,
 528 364(1520), 1049–1058. <https://doi.org/10.1098/rstb.2008.0251>

529 Pol, M. van de, & Verhulst, S. (2006). Age-dependent traits: a new statistical model to separate within- and
 530 between-individual effects. *The American Naturalist*, 167(5), 766–773. <https://doi.org/10.1086/503331>

531 Pugesek, B. H. (1981). Increased reproductive effort with age in the California gull (*Larus californicus*). *Science*,
 532 212(4496), 822–823. <https://doi.org/10.1126/science.212.4496.822>

533 R Core Team. (2021). *R: a language and environment for statistical computing*. Retrieved from R Foundation
 534 for Statistical Computing website: <https://www.R-project.org/>

535 Reichert, S., Berger, V., Jackson, J., Chapman, S. N., Htut, W., Mar, K. U., & Lummaa, V. (2020). Maternal age
 536 at birth shapes offspring life-history trajectory across generations in long-lived Asian elephants. *Journal*
 537 *of Animal Ecology*, 89(4), 996–1007. <https://doi.org/10.1111/1365-2656.13049>

538 Roff, D. A. (2002). *Life history evolution* (2nd ed.). Sunderland, MA: Sinauer Associates.

539 Røgilds, A., Andersen, D. H., Pertoldi, C., Dimitrov, K., & Loeschcke, V. (2005). Maternal and grandmaternal
 540 age effects on developmental instability and wing size in parthenogenetic *Drosophila mercatorum*.
 541 *Biogerontology*, 6(1), 61–69. <https://doi.org/10.1007/s10522-004-7385-8>

542 Schroeder, J., Nakagawa, S., Rees, M., Mannarelli, M.-E., & Burke, T. (2015). Reduced fitness in progeny from
 543 old parents in a natural population. *Proceedings of the National Academy of Sciences*, 112(13), 4021–
 544 4025. <https://doi.org/10.1073/pnas.1422715112>

545 Sellers, K. F., & Premeaux, B. (2020). Conway-Maxwell-Poisson regression models for dispersed count data.
546 *WIREs Computational Statistics*, e1533. <https://doi.org/10.1002/wics.1533>
547 Surget-Groba, Y., Heulin, B., Guillaume, C.-P., Thorpe, R. S., Kupriyanova, L., Vogrin, N., ... Smith, N. (2001).
548 Intraspecific phylogeography of *Lacerta vivipara* and the evolution of viviparity. *Molecular*
549 *Phylogenetics and Evolution*, 18(3), 449–459. <https://doi.org/10.1006/mpev.2000.0896>

550

551 **SUPPORTING INFORMATION**

552 Table S1 - Sample sizes

553 Table S2 - Maternal and grandmaternal age combinations of the 135 litters

554 Figure S1 - M age effects on offspring traits at birth

555 Table S3 - Model selection for M age and GM age effects on survival

556

Table 1. Analysis of grandoffspring body size at birth. We used a mixed-effects model with year of maternal reproduction and maternal identity as random effects. Three outliers were removed to conform to normality assumption of the model. Significant effects are in bold. M age and GM age are maternal age and grandmaternal age, respectively. Estimates are given considering male as the level of reference for grandoffspring sex.

	<i>F</i> -value	df	<i>P</i> -value	Estimate	SE
Intercept				20.74	0.81
M age	6.25	1, 538.3	0.013	0.70	0.28
M age²	8.66	1, 527.5	0.003	-0.08	0.03
GM age	0.14	1, 113.3	0.71	0.12	0.33
GM age ²	0.26	1, 101.9	0.61	-0.02	0.04
grandoffspring sex	8.75	1, 477.3	0.003	-0.60	0.20
M age x grandoffspring sex	0.12	1, 478.3	0.73	-0.02	0.05
GM age x grandoffspring sex	0.03	1, 479.8	0.87	0.01	0.04
M age x GM age	0.08	1, 553.0	0.78	0.01	0.04

Table 2. Analysis of grandoffspring body condition at birth. We used a mixed-effects model with year of maternal reproduction and maternal identity as random effects. Two outliers were removed to conform to normality assumption of the model. Significant effects are in bold. M age and GM age are maternal age and grandmaternal age, respectively. Estimates are given considering male as the level of reference for grandoffspring sex.

	<i>F</i> -value	df	<i>P</i> -value	Estimate	SE
Intercept				-15.68	13.50
M age	2.67	1, 515.0	0.10	9.02	5.00
M age²	6.08	1, 511.8	0.014	-1.11	0.45
GM age	0.38	1, 120.9	0.54	-2.31	5.31
GM age ²	0.19	1, 108.1	0.66	0.27	0.61
grandoffspring sex	9.49	1, 485.4	0.002	11.40	3.70
M age x grandoffspring sex	4.75	1, 583.7	0.030	-2.06	0.94
GM age x grandoffspring sex	5.44	1, 668.8	0.020	-1.89	0.81
M age x GM age	0.22	1, 517.6	0.64	0.30	0.64

Table 3. Analysis of granddaughter litter size. We used a Conway-Maxwell-Poisson regression to take into account underdispersion of data. Significant effects are in bold. M age and GM age are maternal age and grandmaternal age, respectively.

	Z-value	P-value	Estimate	SE
Intercept			-3.72	1.29
M age	0.61	0.54	0.18	0.29
M age ²	-0.42	0.68	-0.02	0.04
GM age	2.18	0.029	0.82	0.38
GM age ²	-1.93	0.054	-0.10	0.05
granddaughter body size	3.92	< 0.001	0.06	0.01
M age x GM age	-0.11	0.91	-0.01	0.07

Table 4. Analysis of granddaughter litter success. We used a logistic regression with quasi-binomial error to take into account overdispersion of data. Significant effects are in bold. M age and GM age are maternal age and grandmaternal age, respectively.

	<i>X</i> ² -value	df	<i>P</i> -value	Estimate	SE
Intercept				-18.80	14.53
M age	0.16	1	0.69	-3.92	10.00
M age ²	0.56	1	0.45	1.54	2.38
GM age	3.97	1	0.046	7.98	5.20
GM age ²	2.14	1	0.14	-0.80	0.56
granddaughter body size	1.38	1	0.24	0.20	0.17
M age x GM age	1.22	1	0.27	-1.01	1.16

Figure legends

Figure 1. Terminology. The individuals of the first generation (F0) are consistently called grandmothers, those of the second generation (F1) mothers and those of the third generation (F2) grandoffspring. The distinction between an age effect, a maternal effect and a grandmaternal effect depends on the number of generations between the individuals and not on the name of the explanatory variable (M age or GM age).

Figure 2. Grandmaternal age effect on grandoffspring body condition at birth. Grandoffspring body condition was dependent on the interaction between GM age and sex as illustrated here in grandoffspring males (blue) and females (red). The figure shows mean values \pm s.e.m., with the predicted relationships of a statistical model including only significant effects.

Figure 3. Grandmaternal age and maternal age effects on grandoffspring survival during their first hibernation. Grandoffspring survival rate was dependent on the additive effects of GM age, M age and sex (Table S3) as illustrated here in grandoffspring females (panel A) and males (panel B). Left panels show the survival rates estimated with a range of 95% confidence interval less than 0.5. Right panels show the range of 95% confidence interval of survival rates for all the combinations of grandmaternal and maternal ages, with the range limit for the value of 0.5 indicated by the dashed line.

Figure 4. Grandmaternal age effects on granddaughter reproduction at two years old. The litter size (panel A) and litter success (panel B) of the two-year-old granddaughters recaptured at their first reproduction depended on grandmaternal age. Litter success is the

proportion of live offspring in a litter. Litter size and litter success are presented as the mean \pm s.e.m., with the predictions of the significant effects of the statistical models (blue lines are predicted values, and shaded areas are their 95% confidence intervals).

Figure 5. Age-dependent multiplication rates of granddaughters. Multiplication rates were computed with age-structured matrix models implemented in the ULM software. This measure takes into account both the effects of grandmaternal age on granddaughter survival and fecundity.

Figure 1

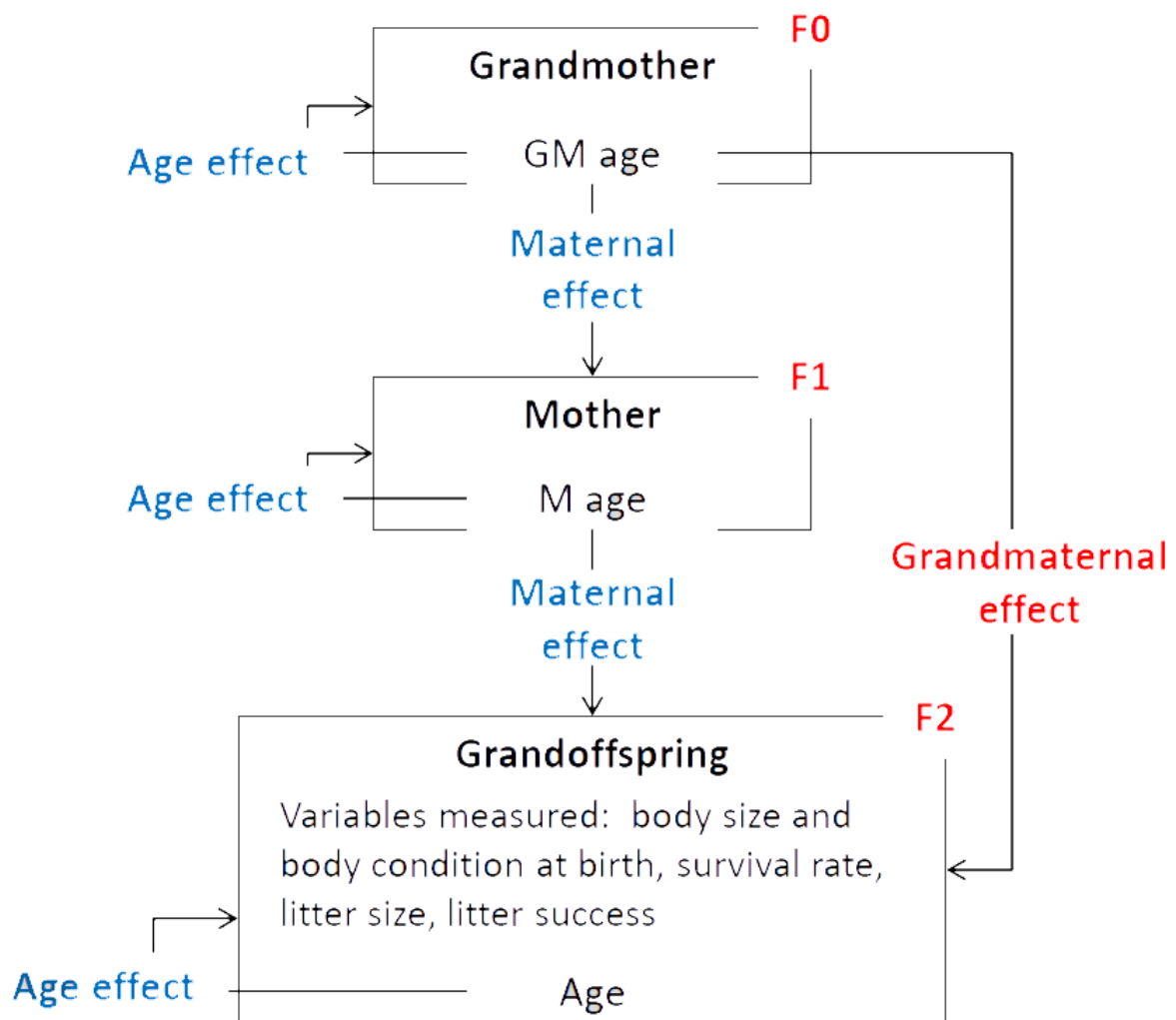


Figure 2

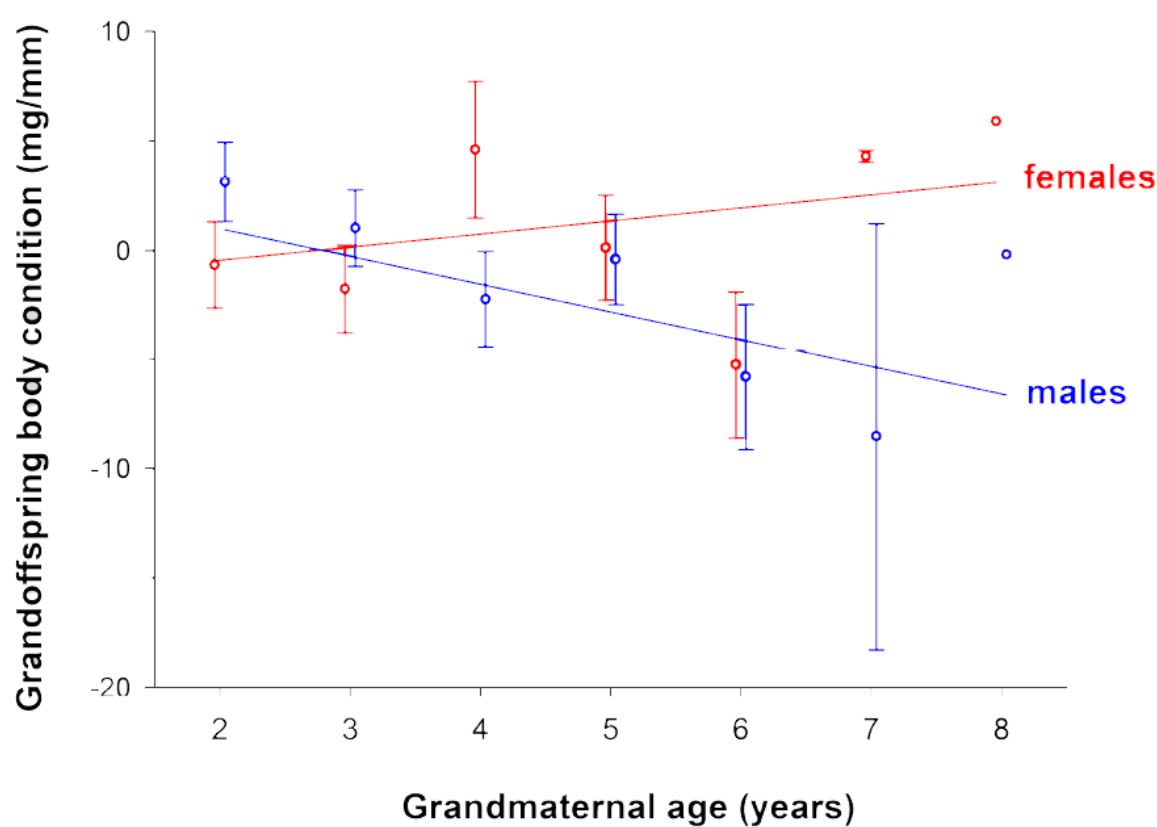


Figure 3

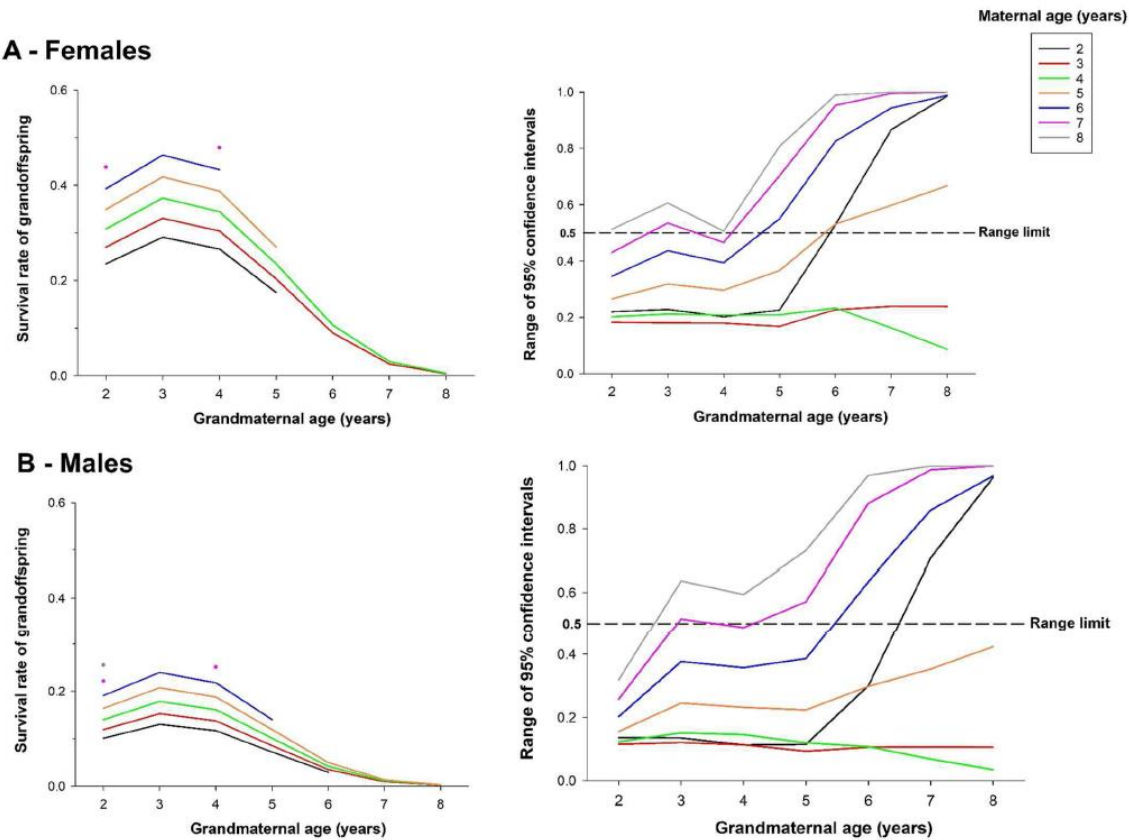


Figure 4

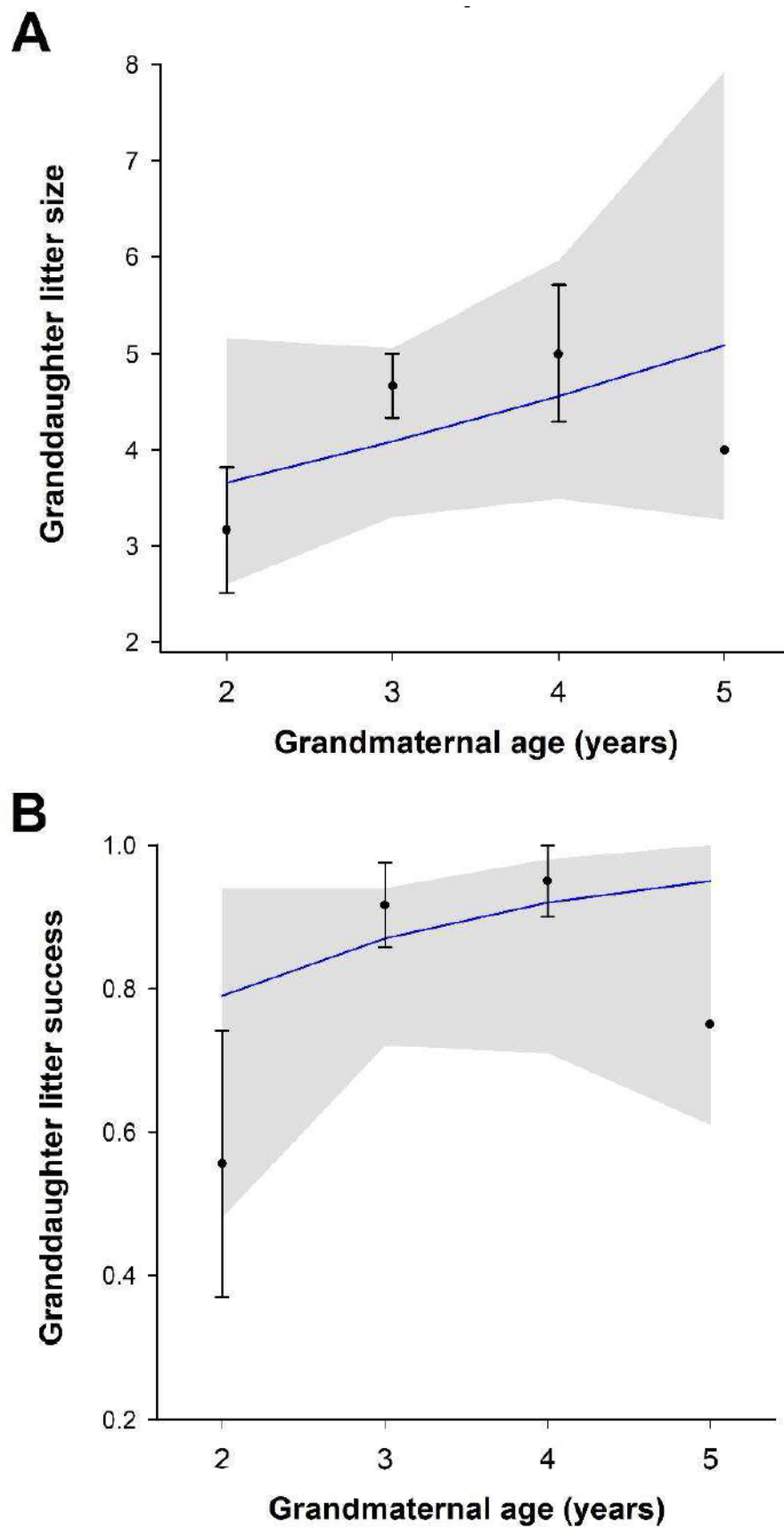


Figure 5

