

Robert Maślak¹, Łukasz Paško¹, Jan Kuszniierz¹,
Magdalena Moska²

**SEX RATIO IN POPULATION OF *LACERTA VIVIPARA*
(JACQUIN, 1787) (SAURIA, LACERTIDAE) IN A LOWLAND
HABITAT OF THE VICINITY OF WROCLAW, SW POLAND**

**STRUKTURA PŁCI W POPULACJI *LACERTA VIVIPARA*
(JACQUIN, 1787) (SAURIA, LACERTIDAE) W NIZINNYM
ŚRODOWISKU OKOLIC WROCLAWIA**

¹*Institute of Zoology, Wrocław University,
Instytut Zoologiczny Uniwersytetu Wrocławskiego*

²*Department of Genetics and Animal Breeding, Wrocław University of Environmental
and Life Sciences,
Katedra Genetyki i Ogólnej Hodowli Zwierząt, Uniwersytet Przyrodniczy we Wrocławiu*

Studies concerning, among others, the sex ratio in a selected population of *Lacerta vivipara* were carried out during four years in the vicinity of Wrocław (SW Poland). The results show that adult sex ratios are male-biased. Assuming prevalence of males among juveniles and yearlings would be burdened with too great error. Reasons for this phenomenon are discussed. The most probable factors determining male-biased sex ratio in studied population seem to be intersexual competition and its impact on female survival and possibly effect of predation. The predation hypothesis is supported by the data on the proportion of males and females with regenerated tails. The proportion is as high as 45.8% in females and only 36.7% in males. This may indicate that females are really more often wounded. Our results confirm that in a lowland populations mean adult mortality is usually lower in males.

KEY WORDS: Lacertidae, *Lacerta vivipara*, *Zootoca vivipara*, sex ratio

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INTRODUCTION

Numerous studies suggest that in lizards unequal sex proportions in populations are common. In many species there are considerable differences in the sex ratios between populations, age classes and particular years. The common lizard (*Lacerta vivipara*) became one of important model organism for several studies focused on reasons and consequences of sex ratio biases (e.g. le Galliard et al. 2003, Uller et al. 2004, Rankin and Kokko 2006, Fitze and le Galliard 2008). Some studies allowed understanding better mechanisms of sexual selection, especially functions of such factors as intra- and intersexual competition (le Galliard et al. 2003, 2005).

This paper shows the estimation of sex ratio in common lizard population and makes an effort to determine the ontogenetic stage on which the factors determining sex ratio operates. The reasons of sex ratio biases with special emphasis on adult sex ratio (ASR) biases are discussed.

MATERIAL AND METHODS

Studies on the ecology of the viviparous lizard (*Lacerta vivipara*) were carried out in during four years in the vicinity of Wrocław (SW Poland). The studied population inhabits a meadow south of the Wrocław district Leśnica. The meadow is located on the left bank of the Bystrzyca River valley, on a higher Holocene terrace (terrace II), at 118 m a.s.l. (Walczak 1970, Szczepankiewicz 1972, 1989). This location makes water inflow difficult. The shape of the meadow is close to rectangular, 250 x 140 m, surrounded by a forest. The vegetation can be classified in the order Molinietalia. Observations in each season started at half of March and were concluded at the beginning of October. Every month the same time was expended on catching of lizards. The lizards were caught mainly in the morning and early afternoon along appointed transects

Lizards were divided on three age group: juveniles, subadults (yearlings) and adults. The sex ratio was studied in every group separately. Notion "juveniles" was used for define specimens born in year of study, "subadults" are lizards after the first winter and "adults", animals which survive at least two winters. All classes are easy to recognize on the basis of the body size.

Sex identification in the studied lizard is not problematic. The most certain characters of sexual dimorphism were taken into account, i.e. thickened tail base in males, where the hemipeneses are situated, and the colouration of the ventral body side. In males, especially in the mating season, there is a bright band usually including 4 median rows of scales. It is orange, ranging from yellow-orange to almost red. The belt is present also outside the mating period, though it is less distinct. Besides, in males there are fine dark spots on the belly. The female's underside is uniformly yellow throughout the year; there are no black spots or they are very few. It should be pointed out, however, that sometimes males are found with a yellow belly (found also in the studied population), as well as females which are yellow-orange underneath. Besides, males have relatively larger heads, but the character is not very much pronounced and thus of no practical significance when determining sex in the field (Dely and Böhme 1984, Juszczak 1987). Described above method of sex recognition concerns of adult animals.

Determining sex of juveniles and one year old individuals based on the above characters is impossible. However, the difference in the number of transverse rows of ventral scales between the sexes can be used. The number of rows is constant throughout

the individual's life. A disadvantage of this method of sex determination is the fact that the character overlaps between the sexes to some degree. The difference in the number of rows of ventral scales in males and females, and the degree of their overlapping vary also between populations (Bauwens and Thoen 1982). For this reason it was necessary to count the rows not only in juvenile and one-year old lizards, but also in adult individuals, in order to ascertain the variability range of the character and the degree of its overlap in the studied population. The rows of ventral scales were counted along the midline of the underside of body, since the course of the rows on the sides of the ventral surface, especially in the region of collare and sc. anale is often disturbed (Lecomte et al. 1992).

In this part of study all caught specimens were used (n=467) and marked toe-clipping method during four years. But for calculation of sex ratio in respective age groups only specimens caught after the May 1st were used. Males appear usually two, three weeks earlier than females after hibernation and are more active in April. They are in this time easier to see and catching. Adopted rule allowed to avoid "artificial" increase in number of males in analysis. Number of animals used for this part of study shows Table 3.

To test hypotheses on the sex ratio in the studied population we used proportion test based on normal distribution within the sample (Łomnicki 1995) and Fisher exact test (StatSoft 2007). In order to find if there is a significant difference between the number of rows of scales between adult males and females, we used a single-factor non-parametric variance analysis (ANOVA) according to Kruskal-Wallis (Łomnicki 1995, StatSoft 2007). The independent variable (factor) is sex, the dependent variable rvs (number of rows of ventral scales). All tests were conducted with the help of Statistica 8 (StatSoft 2008).

RESULTS

The data on the number of rows of ventral scales in males, females, juveniles and yearlings are presented in Figure 1. In the sample of adult lizards males were more numerous (n=88) than females (n=58), the proportion of females in the total sample being 0.40. The variability range of the number of rows of ventral scales in males was 22–28, in females 27–31 (Tab. 1).

Table 1
Tabela 1

Variation in the number of rows of ventral scales in adult lizards
Zmienność w liczbie rzędów tarczki brzusznych w grupie dorosłych jaszczurek

ventral scales rows liczba rzędów tarczki brzusznych	22	23	24	25	26	27	28	29	30	31	32
no of adult males liczba dorosłych samców	2	10	20	37	15	27	10				
no of adult females liczba dorosłych samic						7	19	12	17	3	

The results of Kruskal-Wallis test (Tab. 2) support a statistically significant difference in rvs between adult males and females.

Table 2
Tabela 2Results of Kruskal-Wallis ANOVA
Wyniki testu ANOVA Kruskala-Wallis

Independent variable Zmienne niezależne	No of individuals Liczba osobników	Sum of ranks Suma rang	H Statistics Statystyka H	Significance level Poziom istotności
males samce	121	7640.5	102.69	p = 0.00
females samice	58	8469.5		

Individuals with 27 and 28 rows of ventral scales were omitted when determining sex of juveniles and one years old individuals, since sex determination in such specimens is impossible as they may be either male or female. In the group of one-year old individuals there were 40 such animals (proportion in the total sample = 0.31), and in the group of juvenile individuals 32 (proportion in the total sample = 0.20). In these age classes in the studied sample males prevail as well. In the group of caught one year old specimens there were 49 males and 42 females, among juvenile individuals there were 69 males and 56 females. Proportion of females in the total sample were 0.46 among one-year old individuals and 0.45 among the juveniles.

The results of the proportion test and Fisher exact test are presented in Table 3.

Table 3
Tabela 3Results of Fisher exact test
Wyniki testu dokładnego Fishera

Age classes Klasy wieku	N	m	f	Significance level Poziom istotności	Result of proportion test Wyniki testu dla proporcji
yearlings młode	91	49	42	p = 0,36	p = 0.47
juveniles przezimki	125	69	56	p = 0,24	p = 0.25
adults dorosłe	146	88	58	p = 0,05	p = 0.01

N – no of individuals in a sample, m – no of males in a sample, f – no of females in a sample
N – liczba osobników w próbie, m – liczba samców w próbie, f – liczba samic w próbie

Only in case of one age class (adult individuals) the rejection of hypothesis of the sex ratio 1:1 is justified at $p < 0.05$ (both used tests). Hypothesis of the the population is male-biased should be accepted. In case of yearlings and juvenile individuals accepting this thesis would be burdened with too great an error (though much smaller in the case

of juveniles). It seems likely that the factor determining the prevalence of males in the population acts at the period of maturation or affects adult individuals.

The 95% confidence intervals (95% C.I.) calculated for females and representing possible at $p \leq 0.05$ values of female proportion in the population are: for yearlings – 95% C.I. = [0,36 to 0,57]; for juveniles – 95% C.I. = [0,36 to 0,54]; for adults – 95% C.I. = [0,32 to 0,48].

Obviously, values closest to the middle of these confidence limits are the most probable. This would indicate a superabundance of males among adult individuals about 5 per cent as the most probable.

DISCUSSION

The obtained results indicate that adult sex ratio (ASR) in studied population is male-biased in adult lizards. ASR of natural populations of this species is often female-biased, however geographical and interannual variation occurs, which ranges from 15 to 65% males across years and populations (le Galliard et al. 2005). Below we discuss factors which may be important in sex proportion in populations of this species on different stages of ontogenesis.

In many reptiles it was demonstrated that sex determination was associated with the temperature during the embryonic development (e.g. Bull 1980, 1985, 1987a,b, Warner 1980, Deeming and Ferguson 1988, Janzen and Paukstis 1991). In Lacertidae TSD (temperature dependent sex determination) was described only in *Podarcis pityusensis* and *Podarcis hispanica*. In the latter species the data raise serious doubts (Eichenberger 1981, Janzen and Paukstis 1991).

In *Lacerta vivipara* the sex of birth is determined (at least partly) genetically by a modified ZZ/ZW chromosomal system (Chevalier et al. 1979). Studies in lizards suggest several potential mechanism by which females might adjust their sex-biased investment. In GSD (genetic dependent sex determination) species, mothers could control sex ratio via gamete selection and differential abortion of embryos. This phenomenon might be under environmental control through the impact of gonadotropin and steroid hormones (Krackow 1995, Calsbeek and Sinervo 2004, le Galliard et al. 2005). Females can control the sex ratio by selecting the temperature at which their offspring develop (Robert and Thomson 2001, Shine 2002, Wapstra et al. 2004). Finally, mothers can allocate more resources to one sex biasing the amount of yolk or steroid hormones deposited in the egg (e.g. Olsson and Shine 2001, Painter et al. 2002). Transfer of steroids during egg formation and during embryonic development through a primitive chorioallantoic placenta allow for maternal effect (Gavaud 1986). Heterogametic females (ZW) have the potential to control sex biased investment through preovulation control of chromosome segregation, selective sex-specific abortion of the ovulated follicles, and sex-biased investment into the eggs (Meylan and Clobert 2004, le Galliard et al. 2005).

Studies of Lorenzon et al. (2001) suggest than in the *Lacerta vivipara* the clutch sex ratio at birth varies with environmental conditions. Higher habitat humidity was associated with more male-biased sex ratios at birth. Experimentally increased maternal corticosterone levels led to more female-biased clutches (Meylan and Clobert 2004).

The sex ratio during early stages of ontogenesis have strong impact on fitness and implications for life-history evolution and sex allocation. Studies of Uller et al. (2004) show that females from male-biased clutches grow faster, mature earlier, but have lower

fecundity than females from female-biased clutches. Furthermore, male reproduction was also affected by the sex ratio during embryonic development, with males from male-biased clutches being more likely to successfully reproduce at age one than males from female-biased clutches.

Differences of unequal presence of different sexes on the area may be result of different dispersal strategy. Experiments of le Galliard et al. (2003) demonstrated that maternal presence has a significant effect on sex biased dispersal from natal patch. During the juvenile stage, higher dispersal was found in female offspring in the presence of the mother, whereas males were unaffected. During the yearling stage, the responses of both sexes to the presence of the mother opposed each other. Despite the fact that offspring have been shown experimentally to suffer from competition with adults, competitive interactions may differ between sexes. For example, females tend to compete for resources, whereas males tend to compete for mates (Pilorge 1987, Massot et al. 1992). Thus young males might suffer less from competition with the mother than young females. Additionally, males avoid potential mating with their sisters rather than their mother, and therefore adopt a dispersal strategy than is opposite to that of their sisters (Massot and Clobert 2000).

Reasons for the unequal sex proportions in lizard populations can be sought in differentiated survival rate during hibernation. In the studied species hibernation is necessary to maintain normal reproductive cycle (Gavaud 1983, 1991). Though it requires accumulating adequate energy reserves, and the ability to do this may be higher in males since females are subject to greater energy expenditure because of reproduction, in the viviparous lizard this factor probably does not play a decisive role. It was found that the species was resistant to low temperatures, its hibernation period was shorter than in other lizards of the same latitude, and the survival rate during hibernation was very high and sex-independent (Bauwens 1981).

Survival rate of females that have entered the reproductive phase of life may be lower because of their being more exposed to predation during pregnancy. This is especially important in viviparous species in which all the embryonic development takes place in the female's organism. It can be supposed that the higher mortality of females during that period is one of serious costs paid during the evolution of viviparity (e.g. Shine and Bull 1979). Some studies indicate that pregnant females are to some degree handicapped when a quick escape from danger is necessary. Their speed is lower and they are less eager to run. The increased weight of female in that period to a considerable degree affects her behaviour, among others her ability to quick escape and avoiding the danger from predators (Bauwens and Thoen 1981, Damme et al. 1989). Some authors question results of such studies. According to Heulin (1984) pregnant females are not so much exposed to predators, since their individual ranges are smaller than in males and non-pregnant females, and thus they always stay close to their hiding places. Lecomte et al. (1993) do not share the opinion that pregnant females, when in danger, start their escape with delay. They point additionally to the fact that the behavioural differences are strongly associated with ambient temperature. Pregnant females of *Lacerta vivipara* spend less time in the sun than non-pregnant females, which these authors associate not so much with protection against predators, as with the necessity to ensure proper temperature for the embryos.

The adult sex ratio (ASR) is important parameter of population demography. A prevalent hypothesis is that sex proportion by adults are regulated by intrasexual competi-

tion, which would cause more mortality or emigration in the sex of increasing frequency (Fisher 1930, Greenwood 1980, Clutton-Brock et al. 2002). Le Galliard et al. (2005) published results of their work which shows opposite effect. They conducted long term study of the common lizard in semi-natural conditions and manipulated sex proportion in particular populations. Male mortality and emigration are not higher under male biased adult sex ratio. The absence of difference in male mortality between female-biased and male-biased populations contrasted sharply with strong age-dependent differences in female mortality. The mortality of yearlings and adult females was increased dramatically by male excess, whereas the survival of juvenile females was not significantly affected. Considering impact of skewed ASR on female reproductive output, Le Galliard et al. (2005) found that the proportion of gravid females was similar between treatments, but fecundity (number of offspring that successfully hatched) was lower in male-biased populations. Thus, male excess reduced population growth by decreasing both female survival and recruitment. The negative impact of male excess on female survival and reproduction could arise from intersexual competition and/or male aggression during mating (Smuts and Smuts 1993, Magurran and Seghers 1994). The latter includes direct physical effects (males bite females during mating) (Bauwens and Verheyen 1985), stress (induced by male mating attempts) that inhibits reproduction (More and Jessop 2003), or loss of energy due to sexual harassment by males (Magurran and Seghers 1994). In the studies of Le Galliard et al. (2005) the most important role played male aggression. The number of mating scars and wounds on females due to males' biting during mating attempts were 2- to 3-fold more frequent in male biased populations. The female death happened mainly during the spring mating season. The model with stochastic sex-ratio fluctuations constructed by Le Galliard et al. (2005) to estimate how such results would affect population persistence, revealed that the extinction risk owing to such stochasticity increased as a result of male aggression: incorporating male aggression in a model of an isolated population led probable extinction within 40 years (Rankin and Kokko 2006). Some studies show that by common lizard which is species with high female mating costs, positive directional sexual selection on male size was weaker in male-biased populations than in female-biased (Fitze and Le Galliard 2008).

In case of our study male-biased adult sex ratio (ASR) seems to be result of some factors described above. We should exclude mechanisms acting during embryonic development (such as TSD, gamete selection and differential abortion of embryos) and during early stages after hatching. Thus the most probable factors determining male-biased sex ratio seem to be intersexual competition and its impact on female survival and possibly effect of predation. Predators of lizards on the study area were recognized (Mašlak and Paško 1999). This is supported by the data on the proportion of males and females with regenerated tails in the studied population. The proportion is as high as 45.8% in females and only 36.7% in males. This may indicate that females are really more often disturbed and subject to damage. Our results confirm that in a lowland populations mean adult mortality is usually lower in males (Massot et al. 1992, Heulin et al. 1997).

The method of determining sex of juvenile individuals and yearlings requires a separate discussion. It was possible to ascertain the sex of 79.6% juveniles and 69.5% yearlings, and in males the number of rows of ventral scales was 22–28, in females 27–31. In the population studied by Boulenger (1920) males had 21–27 rows of ventral scales,

females 24–31. According to Wermuth (1955) males have 21–30 and females 25–34 rows, but 2 should be subtracted from each value obtained by this author, since he included 2 rows of preanal scales in ventral scales. Bauwens and Thoen (1982) found 23–28 rows in males and 27–34 in females (sex determination possible in 75% cases), and in another studied population 24–29 rows in males and 27–31 in females (sex determination possible in 60% cases). Some studies indicate existence of populations where the overlap in this character between the sexes is so narrow that sex can be determined even in over 90% cases (Lecomte et al. 1992). Studies on such populations would allow for a better recognition of sex structure in various age classes.

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(SAURIA, LACERTIDAE) W NIZINNYM ŚRODOWISKU
OKOLIC WROCŁAWIA**

S t r e s z c z e n i e

Badania dotyczące m.in. struktury płci przeprowadzono w ciągu czterech lat na wybranej populacji jaszczurki żyworodnej, *Lacerta vivipara*, w okolicy Wrocławia. Wyniki wskazują, że w populacji występuje przewaga samców w populacji dorosłych osobników. W przypadku młodych i jaszczurek po pierwszym zimowaniu – nie stwierdzono istotnych statystycznie różnic w strukturze płci. Dyskutowane są przyczyny tego zjawiska. Najbardziej prawdopodobnym czynnikiem decydującym o przewadze samców u dorosłych zwierząt jest konkurencja między samcami i jej wpływ na przeżywalność samic oraz możliwy wpływ drapieżnictwa. Hipoteza związana z tym ostatnim zjawiskiem znajduje uzasadnienie w danych dotyczących proporcji samic i samców z regenerowanymi ogonami. Takie samice stanowią 45,8%, a samce tylko 36,7. Może to oznaczać, że samice są bardziej narażone na drapieżnictwo. Uzyskane rezultaty potwierdzają, że w populacjach nizinnych średnia śmiertelność dorosłych samców jest mniejsza niż samic.

SŁOWA KLUCZOWE: Lacertidae, *Lacerta vivipara*, *Zootoca vivipara*, proporcje płci

Reviewer – Recenzent: Prof. Dr. Sci. Władysław Zamachowski, Pedagogical University of Cracow