BODY SIZE VARIATION IN *RANA TEMPORARIA* POPULATIONS INHABITING EXTREME ENVIRONMENTS

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Abstract: We studied the variation in body size in populations of a widespread anuran species, *Rana temporaria*, from high altitude and latitudes. Our results indicated a variable interannual pattern of body size, suggesting that body size in extreme environments is influenced by many factors. This indicates that long-term series of observations are needed to separate natural fluctuations from man-induced changes.

Keywords: *Rana temporaria*, extreme environments, body size, interannual variation

1. Introduction

During the last decades, many amphibian species have declined from high altitude area, even in habitats apparently without human impact [1, 2]. The causes of some declines remain unknown. Understanding of the life history characteristics of the amphibian populations that inhabit extreme environments at high altitude and latitude is an important step in the evaluation process of the potential causes of decline. Genetic and environmental factors (e.g. temperature, rainfall, trophic resources, competition, predators) determine variation in the life history traits of species occupying a large geographic area [3]. Low temperature, associated with high altitude/latitude, reduces the activity period and the time available for resource exploitation [4]. Temperature affects the duration of hatching and metamorphosis in amphibians. The increase in the adult body size has been frequently associated with a cold annual temperature [5, 6]. Most studies of variation in amphibians body size have focused on latitudinal and altitudinal variation, e.g. trying to establish if the amphibian species follow the Bergmann's rule [7, 8]. Studies on the interannual variations in amphibians body size generally analyze difference in body condition [9, 10], or variation in age and size at maturity [6].

The Common Frog (*Rana temporaria*) is the most widespread amphibian species in Europe [11]. Its distribution reaches 71^{o} N in Fennoscandia [12] and it can be found even at altitudes of 2600 m [12]. The wide altitudinal and latitudinal range of this species, allows comparisons of life-history traits over a broad range of conditions. In a previous publication we analyzed the altitudinal and latitudinal body size variation among populations from high altitude and latitude of *R. temporaria* testing if the variation pattern is according to the Bergmann's rule [13]. In this study we analyzed interannual body size variation in the same *Rana temporaria* populations, in order to evaluate if the pattern of variation in body size changes in time. We tested the following predictions: i) there is no interannual variation in body size and ii) the mean body size of the frog populations from subarctic regions shows significant variation during a growth season.

2. Material and Methods

R. temporaria populations were studied from Kilpisjarvi, Finland (latitude N 69^{o}) in 2003 (August) and 2009 (July), Kolari, Finland (latitude 67.2^{o}) in 2009 (July) and in Retezat National Park, Romania (latitude N 45^{o}) in 2004 (September) and 2009 (August). Latitude and altitude were recorded for each population by using a handheld Garmin GPS.

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Captured individuals were sexed, weighed (W) to the nearest 0.01 g with a portable electronic balance (AccuLab Pocket Pro), and snout-vent length (SVL) was measured to the nearest 0.5 mm with dial-calipers. Data were log transformed prior to analyses. For comparisons between years and sites we used One-way analysis of variance (ANOVA) and Analysis of covariance (ANCOVA) to compare the slopes of the regression lines. Statistical analyses were performed using SPSS ver. 10.0 (SPSS Inc., 1999).

3. Results and Discussions

A total of 347 individuals were measured and weighed in 2003/2004, of which 237 juveniles and 110 adults (67 males and 43 females) and 157 individuals in 2009 (66 juveniles, 43 females and 48 males). Both log transformed W and SVL were normally distributed (W: $D = 5.06$, $p < 0.001$; SVL: $D = 3.75$, $p < 0.001$). The body size indices of the studied populations are presented in Tables 1 and 2. There was no significant difference in SVL between Retezat National Park and Finland - Kilisjarvi populations. We found significant differences in the mean body size indices between the two stations from Finland (Table 3).

We then compared W and SVL from different years for the same population. We found significant differences in the interannual variation of the body size indices for juveniles in both Finland and Retezat populations, and in the mean weight for females. Males showed only in Retezat a significant interannual variation in the body size indices (Table 4). We also compared the slopes of the regression lines of W as a function of SVL. The slopes of the regression lines are significantly different for all adults in Retezat and Finland (Fig. 1: $F_{1,56}= 81.41$, $p<0.001$; Fig. 2: $F_{1,27}=102.5$, $p<0.001$; Fig. 3: $F_{1,41}=58.3$, $p<0.001$; Fig.4: $F_{1,48}=48.4$, $p<0.001$; Fig. 5: $F_{1,28}=28.8$, $p<0.001$; Fig. 6: $F_{1,24}=43.1$, $p<0.001$).

Table 1. Snout-vent length (SVL) variation according to sex and age classes, in populations of *R. temporaria* from Finland and Romania in 2009 (FN = Finland - Kilpisjarvi; FS = Finland - Kolari, RNP = Retezat high altitude; SD = standard deviation; Min = minimum; Max = maximum).

Populations	Females			Males			Juvenils		
	Average	SD	Min-Max	Average	SD	Min-Max	Average	SD	Min-Max
FS	71.24	6.00	62 - 85.6	69.89	4.45	63.4 - 75	42.17	11.28	28.3 – 59.7
FN	61.15	4.15	55.1 - 67.9	62.46	3.37	57.8 - 70.6	45.95	6.95	24.2 -55.5
RNP	76.78	10.29	55.7-91.8	73.41	7.41	55.6 - 84.7	44.80	12.30	27.8 -59.8

Table 2. Weight (W) variation according to sex and age classes, in populations of *R. temporaria* from Finland and Romania in 2009 (FN = Finland - Kilpisjarvi; FS = Finland - Kolari, RNP = Retezat high altitude; SD = standard deviation; Min = minimum; Max = maximum).

Populations	Females			Males			Juvenils		
	Average	SD	Min-Max	Average	SD	Min-Max	Average	SD	Min-Max
FS	18.96	5.17	10.5 - 27.3	17.27	3.18	12.6-22.9	4.33	3.27	1.1 - 10.4
FN	14.31	4.91	9.1 - 24.8	15.85	4.34	10 - 24.2	7.78	2.85	1-13.2
RNP	34.69	12.49	14.07-57.15	36.20	11.40	13.78 -59.2	9.28	5.86	1.7 - 15.04

Table 3. Comparison of SVL and W between the three *R. temporaria* populations, by using ANCOVA (FN = Finland - Kilpisjarvi; FS = Finland - Kolari, RNP = Retezat high altitude; N = sample size, *P < 0.05, ***P < 0.001, NS = not significant).

			W			SVL		
	N		Average		Fa	Average		Fa
FN vs FS	FN	FS	FS	FN		FS	FN	
Female	13	16	18.956	14.308	6.069*	71.244	61.154	26.384***
Male	17	9	17.267	15.853	0.737^{NS}	69.89	62.46	22.88***
Juveniles	50	9	4.333	7.779	10.381*	42.17	45.95	1.748^{NS}
FN vs RNP	FN	RNP	FN	RNP		FN	RNP	
Female	13	14	14.308	34.69	19.504***	61.154	76.78	3.627^{NS}
Male	17	22	15.853	36.20	17.93***	62.46	73.41	1.778^{NS}
Juveniles	50	7	7.779	9.28	5.903*	45.95	44.8	0.198^{NS}

Table 4. Comparison of the interannual variation in SVL and W between the three *R. temporaria* populations, by using ANCOVA (FN = Finland - Kilpisjarvi; FS = Finland - Kolari, RNP = Retezat high altitude; N = sample size, *P < 0.05, ***P < 0.001, NS = not significant).

			W			SVL		
	N		Average		Fa	Average		Fa
FN 2003 vs. 2009	2003	2009	2003	2009		2003	2009	
Female	29	13	31	14.308	4.235*	65.94	61.154	3.70^{NS}
Male	32	17	31.76	15.853	3.713^{NS}	67.59	62.46	0.43^{NS}
Juveniles	197	50	3.17	7.779	78.41***	29.01	45.95	120.01***
RNP 2004 vs. 2009	2004	2009	2004	2009		2004	2009	
Female	14	14	61.16	34.69	14.56***	82.7	76.78	2.24^{NS}
Male	35	22	47.14	36.20	12.21***	77.07	73.41	4.49*
Juveniles	40	7	2.19	9.28	22.56***	24.42	44.8	19.4***

The variation in the adult body size reported in amphibians can be induced by several factors, including genetic and environmental differences, such as: duration of the activity period, food availability and climatic conditions [6, 14]. Laugen et al. (2005) found that body size decreased with latitude in the Scandinavian Common Frog populations. Comparisons between populations from Western Europe with different activity periods report increases in the mean length, as activity period gets shorter [6]. *Rana temporaria* populations from the analyzed area show a variable pattern in weight and length. Băncilă et al. (2010) found that latitudinal and altitudinal variation patterns in juvenile body size were according to the Bergmann's rule. We found an opposite pattern for juveniles, with decreases in the body size as the activity period gets shorter. Since juveniles have higher growth and development rate than adults, difference could be observed even in the case of short periods of time between sampling.

Interpopulational variation in the adult body size could be caused by differences in the age structure. Growth rates in amphibian species can dramatically decrease after the attainment of sexual maturity (e.g. Miaud et al. 1999). Thus, delayed reproduction can allow a prolonged growth period and the attainment of a larger adult size.

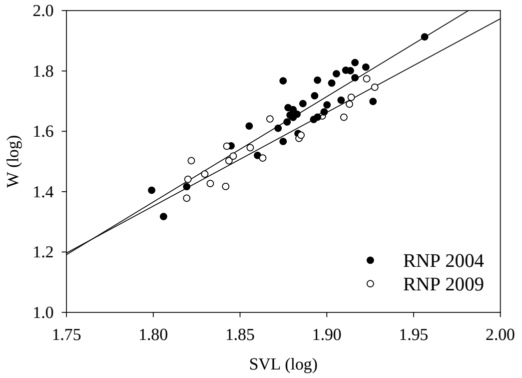

Fig. 1. Body size indices for males in RNP populations, 2004 (N=35; R^2 = 0.72) and 2009 (N=22; R^2 = 0.90).

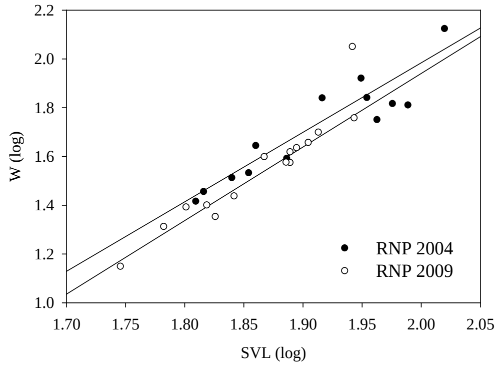

Fig. 2. Body size indices for females in RNP populations, 2004 (N=14; R^2 = 0.75) and 2009 (N=14; R^2 = 0.95).

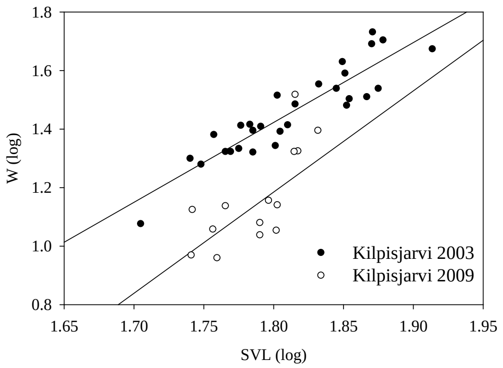

Fig. 3. Body size indices for females in Finland-Kilpisjarvi, 2003 (N=29; R^2 = 0.63) and 2009 (N=13; R^2 = 0.59).

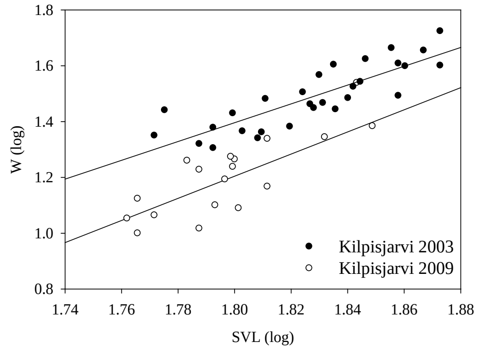

Fig. 4. Body size indices for males in Finland-Kilpisjarvi, 2003 (N=32; R^2 = 0.70) and 2009 (N=17; R^2 = 0.63).

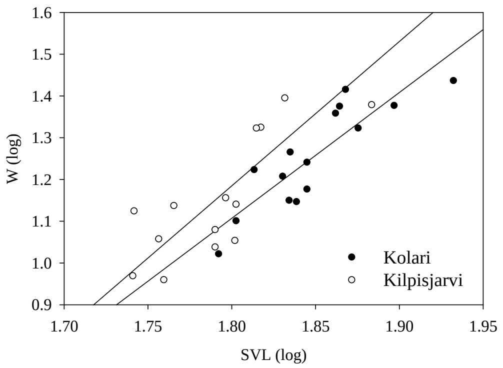

Fig. 5. Body size indices for females in Finland Kilspijarvi (N=13; R^2 = 0.59) and Finland Kolari 2009 (N=16; R^2 = 0.71).

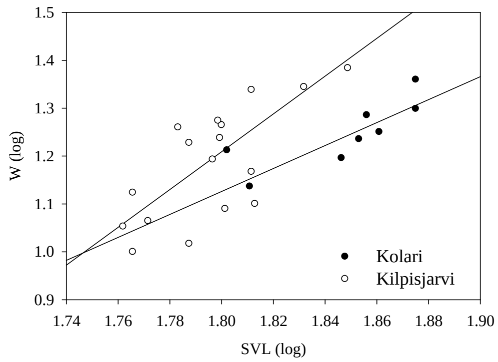

Fig. 6. Body size indices for males in Finland Kilpisjarvi (N=17; R^2 = 0.89) and Finland Kolari 2009 (N=9; R^2 = 0.70).

Factors such as temperature and humidity can directly affect the activity period and the availability of food, influencing the growth rate and the fat stores; hence they could consequently determine significant interannual variation in the body size. Populations from both analyzed areas exhibit interannual variation in weight and length. This variation mainly affects the weight and could be the result of the differences in the sampling period. The pattern of the adult size variation could also directly result from the variation in the population age structure. Further analyses are necessary to determine whether variation in the age structure are contributing or not to the interannual body size indices. Results suggest that many factors affect the body size in extreme environment and long-term series of observations are needed in order to separate natural fluctuations from the human impact/global warming.

4. Conclusions

This study stresses the importance of analyzing interannual variation of life history traits, because one-year data may not properly reflect the features of a population and this issue becomes important in the context of global changes and their possible effects on the amphibian populations.

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