

Inverse Rensch's rule in a frog with female-biased sexual size dimorphism

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Abstract Rensch's rule claims that sexual size dimorphism (SSD) increases with body size when males are larger but decreases with body size when males are smaller. Chinese wood frog *Rana chensinensis* is a medium-sized species with female-biased size dimorphism. Using data on body size and age in 27 populations covering the full known size range of the species, we tested the consistency of allometric relationships between the sexes with Rensch's rule and evaluated the hypothesis that SSD is largely a function of age differences between the sexes. The results showed that level of female-biased SSD increased with increasing mean size, supporting the inverse of Rensch's rule. Moreover, most of the variation in SSD can be explained in terms of differences in age between the sexes in populations.

Keywords Age difference · Sexual size dimorphism · *Rana chensinensis* · Rensch's rule

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Introduction

Sexual size dimorphism (SSD) occurs when males and females of a species or population differ systematically in body size (Fairbairn et al. 2007). SSD can be explained by several specific hypotheses (Andersson 1994; Dale et al. 2007). More general is empirical Rensch's rule (Abouheif and Fairbairn 1997) claiming that SSD increases with overall body size across species where males are larger the sex and decreases with body size where females are larger. The Rensch's rule is a pervasive macroecological pattern that has been observed in various animal taxa (Colwell 2000; Székely et al. 2004; Dale et al. 2007).

The degree of SSD within species is usually viewed as resulting from sex-specific equilibrium of sexual, fecundity, and natural selections (Colwell 2000; Blanckenhorn 2005) and the correlational selection (Fairbairn 1997). As the most supported hypothesis, sexual selection predicts that intense selection drives the evolution of body size of the selected sex, usually the males, with weaker correlated selection on body size in the other sex (Dale et al. 2007). Moreover, life-history hypothesis predicts that difference in development time (age) and growth between males and females also results in extent of SSD (Monnet and Cherry 2002; Blanckenhorn et al. 2007). Sexual selection is an ultimate cause of the extent of SSD, while growth rate and age are proximate causes.

Studies of Rensch's rule, as far as we are aware, focused mainly on animal populations in the laboratory with the inclusion of insects (Fairbairn et al. 2007), domestic goats and sheep (Polák and Frynta 2009), domestic chickens (Remeš and Székely 2010), and cattle breeds (Polák and Frynta 2010). Recently, researchers are increasingly interested to extend Rensch's rule to vertebrates in their natural environment intraspecific level (Lengkeek et al. 2008;

Herczeg et al. 2010). However, intraspecific study of Rensch's rule in frogs is unavailable. In this study, we provided one of the first intraspecific tests of Rensch's rule in frogs, using data on SSD from 27 (Table 1) Chinese Wood frog *Rana chensinensis* populations differing markedly in their mean body size and covering the known size range of the species. We tested the consistency of allometric relationships between male and female body size with Rensch's rule; we also evaluated the plausibility of the hypothesis that population divergence in SSD was largely a function of age differences between the sexes.

Materials and methods

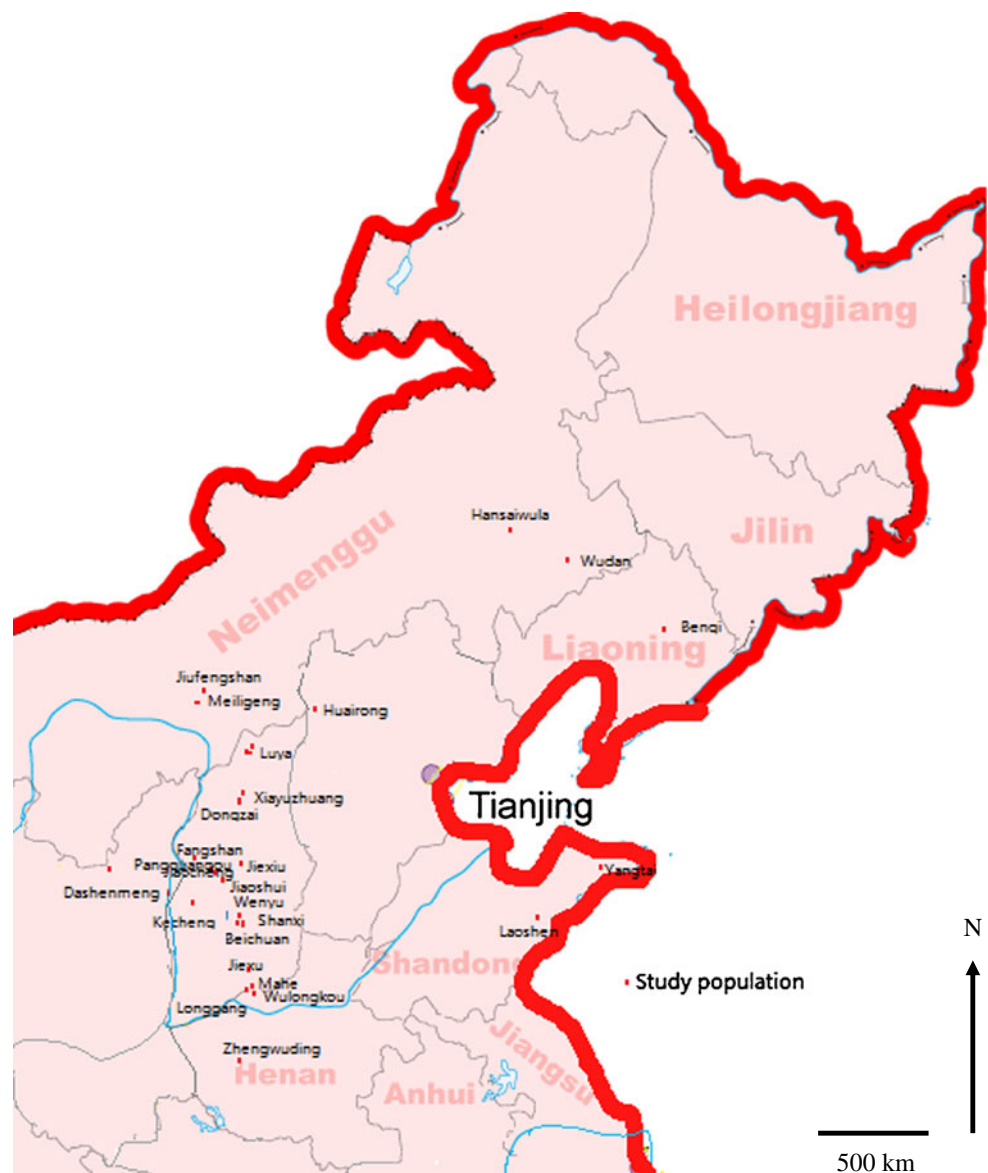
Chinese Wood frog *R. chensinensis* is a medium-sized species with female-biased size dimorphism (Xie et al. 2000). The species is adapted to a wide range of climate regimes and is found over a broad geographical area in terms of both latitude

(between 31 °N and 44 °N) and elevation (300–3,100 m) (Xie et al. 2000). Life-history characteristics along environmental gradients have been investigated in the species in recent years (Lu et al. 2006; Ma et al. 2009; Chen et al. 2011a, b). From these literatures, we obtained morphological data on mean age and body size from 27 populations with female-biased SSD (Table 1). Each population was not equidistant from the other, and there was no direct gene flow between populations (Fig. 1). All the individuals used in this study were adults, based on direct observation of the secondary sexual characteristics (i.e., the vocal sacs in adult males and the eggs readily visible by the skin of the abdomen in adult females). Each individual body size (the snout–vent length, SVL) was measured using a caliper by holding it in normal posture. Individual age was estimated by skeletochronology because this method has been successfully used in assessing age for most anurans based on lines of arrested growth recorded in cross sections of long bones in phalanges (Liao and Lu 2011). The data from literature were reliable because the same standard

Table 1 Location, altitude, sample size (males/females), mean body size, and age in males and females of 27 populations and references of published papers by Lu et al. (2006); Ma et al. (2009); Chen et al. (2011a, b)

Sites	Location	Altitude (m)	Sample size	Males SVL	Females SVL	Males age	Females age	References
Luya mountain	39°40' N, 112°20' E	1,400	27/21	39.5	45.0	1.9	2.7	Lu et al.(2006)
Luya mountain	39°40' N, 112°20' E	1,700	22/20	42.5	44.0	2.1	2.6	Lu et al.(2006)
Luya mountain	39°40' N, 112°20' E	2,000	24/20	43.5	50.5	2.4	3.9	Lu et al.(2006)
Beichuan	36°34' N, 112°46' E	567	53/33	41.8	43.7	1.4	1.7	Ma et al. (2009)
Shanxi	36°35' N, 112°54' E	1,470	38/14	42.9	50.5	1.3	3.4	Ma et al. (2009)
Wenyu	37°48' N, 112°27' E	1,700	25/21	47.0	52.5	2.0	2.8	Ma et al. (2009)
Benqi	41°04' N, 124°01' E	367	63/60	60.6	67.7	2.2	2.4	Chen et al. (2011a)
Zhengwuding	33°40' N, 112°18' E	1,041	10/9	39.3	50.3	2.9	3.6	Chen et al. (2011b)
Wulongkou	35°12' N, 112°42' E	261	11/4	48.6	56.0	3.0	3.3	Chen et al. (2011b)
Manghe	35°18' N, 112°24' E	845	3/3	44.4	45.8	3.3	3.0	Chen et al. (2011b)
Longgang	35°43' N, 112°02' E	1,169	19/13	37.9	38.2	2.9	1.8	Chen et al. (2011b)
Dashanmeng	35°51' N, 108°18' E	1,172	5/10	40.4	42.7	2.6	3.0	Chen et al. (2011b)
Laoshen	36°12' N, 120°36' E	273	16/12	44.7	45.3	2.9	2.7	Chen et al. (2011b)
Kecheng	36°35' N, 111°14' E	1,365	25/15	39.7	42.1	2.8	3.1	Chen et al. (2011b)
Jiexiu	37°04' N, 112°04' E	815	19/18	39.5	40.2			Chen et al. (2011b)
Fangshan	37°11' N, 111°14' E	1,345	15/20	39.3	41.9	2.7	2.8	Chen et al. (2011b)
Yantai	37°18' N, 121°44' E	112	20/10	40.7	44.8	2.9	3.6	Chen et al. (2011b)
Jiaoshui	37°31' N, 112°04' E	770	3/3	37.2	44.3	2.0	3.0	Chen et al. (2011b)
Jiaocheng	37°40' N, 111°48' E	1,257	27/22	39.8	41.3	2.7	2.9	Chen et al. (2011b)
Panguangou	37°50' N, 111°29' E	1,180	18/15	46.1	51.0	3.3	4.1	Chen et al. (2011b)
Dongzhai	38°49' N, 112°05' E	1,588	14/12	39.0	42.1	3.1	3.6	Chen et al. (2011b)
Xiayuzhuang	38°55' N, 112°13' E	1,620	13/8	40.6	43.5	3.0	3.1	Chen et al. (2011b)
Huairong	40°38' N, 114°31' E	991	9/20	40.4	44.5	3.1	3.2	Chen et al. (2011b)
Meiligeng	40°40' N, 109°26' E	1,181	9/9	41.3	48.3	2.7	3.6	Chen et al. (2011b)
Jiufengshan	40°43' N, 110°41' E	1,321	42/8	38.9	46.8	2.9	3.8	Chen et al. (2011b)
Wudan	43°03' N, 119°16' E	546	11/5	41.7	46.1	2.7	3.8	Chen et al. (2011b)
Hansaiwula	44°15' N, 118°38' E	973	8/6	40.1	48.5	3.6	3.5	Chen et al. (2011b)

Fig. 1 Topographic map showing the location of the sampling of 27 populations in northern China



measurements were performed as mentioned, and all individuals were deposited in a standard museum for all populations. Following a protocol of Monnet and Cherry (2002), we calculated SSD as $\log(\text{female mean size}) - \log(\text{male mean size})$ ratio and sexual age difference (SAD) as $\log(\text{female mean age}) - \log(\text{male mean age})$ difference for each population.

Body size and age were \log_{10} -transformed to achieve normality. We applied a general linear model with \log_{10} (SVL) as dependent variable and population and sex as fixed factors to test for sex differences in mean SVL among the 27 populations. The standard ordinary least squares (model I) regression would be statistically incorrect because males' size or females' size measurements are fixed and both are measured with error. To test for Rensch's rule, we fitted major axis regression (model II) of \log_{10} (female size) against \log_{10} (male size) using the population means, and we tested the null hypothesis of $\beta = 1$ using 95 % confidence

intervals (Fairbairn 1997). Model II regression was done manually following that of Sokal and Rohlf (1981). We performed Pearson correlation to test the effect of SAD on the degree of SSD. All statistical tests were two-tailed and were conducted by using Type III sums of squares in SPSS 17.0 software.

Results

The degree of SSD differed significantly among populations (population: $F_{23,53} = 68.055$, $P < 0.001$; sex: $F_{1,53} = 11.055$, $P < 0.001$; and population \times sex: $F_{23,53} = 9.524$, $P < 0.001$). Model I regression showed a significant relationship between male size and female size across populations, and the relationship was significantly different from 1 ($F_{1,26} = 62.746$, $R^2 = 0.715$, $\beta = 0.716$, $SE(\beta) = 0.090$, $P < 0.001$; Fig. 2). Model II

regression revealed a close slope and is indicative of a similar relationship ($\beta=0.846$, $SE(\beta)=0.106$), confirming that variation in SSD supported an inverse of Rensch's rule.

As SSD and SAD had a normal distribution (one-sample Kolmogorov–Smirnov two-tailed test: SSD $Z=0.602$, $P=0.861$; SAD $Z=0.781$, $P=0.657$), we were able to perform Pearson correlation analysis to test the relationship between SAD and the degree of SSD, which also indicated a highly significant relationship across populations ($r=0.530$, $P=0.005$, $n=26$). Plots of SSD as a function of SAD were shown in Fig. 3.

Discussion

Most anuran species exhibit female-biased SSD (Shine 1979). Sexual selection, fecundity selection, and sexual age difference can explain this pattern of size dimorphism (Shine 1979; Monnet and Cherry 2002; Herczeg et al. 2010). For *R. chensinensis*, the relationship between SSD and body size showed the inverse of Rensch's rule. In other words, female-biased SSD displayed a hyperallometric relationship with the mean body size such that SSD increased with increasing mean body size. Similar results are observed previously in other studies (Abouheif and Fairbairn 1997; Fairbairn 1997; Colwell 2000; Herczeg et al. 2010). The inverse Rensch's rule in *R. chensinensis* suggested that the most important evolutionary point which fecundity selection acted increases reproductive output in large females, a phenomenon proven both within and among populations (Lu 1994).

Although studies of Rensch's rule is originally framed for interspecific comparisons, its predictions can be also tested

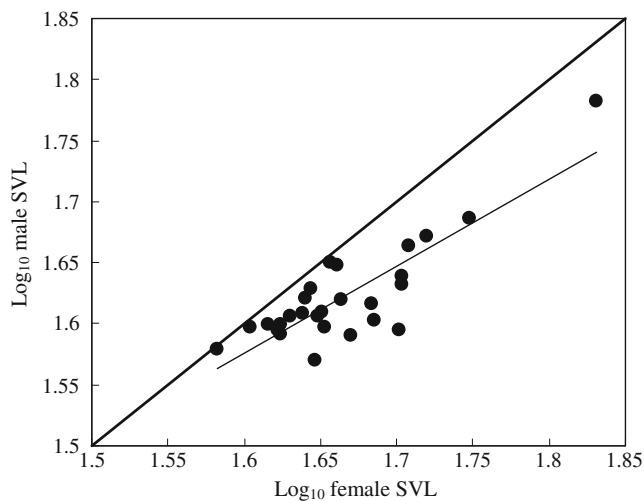


Fig. 2 Allometry of sexual size dimorphism in *Rana chensinensis*. Linear (model I) regression line ($\beta=0.716$) with 95 % confidence interval (dotted line) is shown. The thick gray line represents isometry, i.e., $\beta=1$. Each dot represents a single population based on the mean body size of males and females

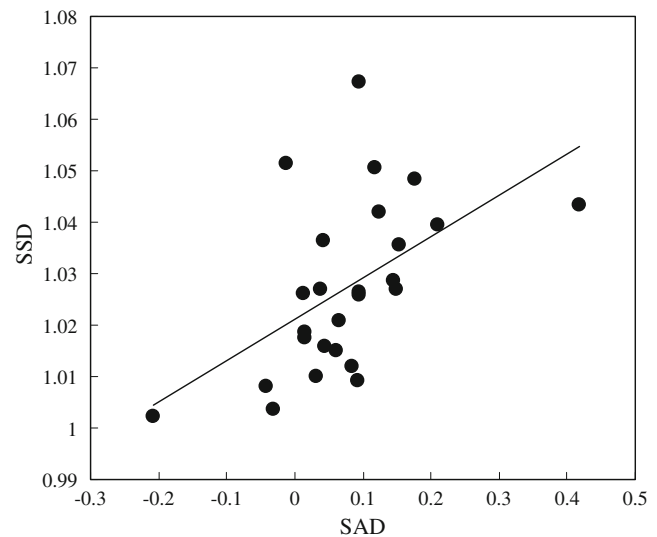


Fig. 3 SSD (log (female mean size)–log (male mean size) ratio) as a function of SAD (log (female mean age)–log (male mean age) difference) in *R. chensinensis* populations. Each dot represents a single population

among populations within species where the mechanisms behind SSD allometry can be explained by detailed knowledge about the species' biology (Colwell 2000; Székely et al. 2004; Blanckenhorn et al. 2007). For *R. chensinensis*, body size/SSD trends from the wild can be interesting; phenotypic plasticity may be the cause behind the observed patterns because environmental condition influences markedly the expression of SSD (Lu et al. 2006). Moreover, genetic basis can explain the inverse of Rensch's rule in nine-spined stickleback *Pungitius pungitius* based on the common garden experiment (Herczeg et al. 2010). Population genetic differentiation might result in the inverse of Rensch's rule because of the data on body size and age in *R. chensinensis* resulting from over a broad geographical area. Solving the question by common garden experiment in the future is necessary.

For ectothermic indeterminate growers like anurans, it has been shown that body size differences can be explained by age differences between sexes (Monnet and Cherry 2002; Liao and Lu 2010a, b; Liao and Lu 2011; Liao et al. 2010, 2011; Liao 2011; Liao and Lu 2012). Comparisons across populations using standard techniques of correlation showed a highly significant correlation between SSD and SAD. Thus, in the populations we have considered, SAD is one mechanism that most probably contributes to the extent of SSD across populations. Moreover, SSD can result from differences in growth rate and/or time available for growth, and these two factors can act simultaneously.

Some reservations may be propitious to our methodology. First, population mean age and size in this species is unstable because of the small sample in some populations. Second, skeletochronological assessment of an individual's age might

be problematic because resorption of the first lines and the presence of double or false lines affect the accuracy in estimating age (Hemelaar 1981). However, Monnet and Cherry (2002) found that if there is systematic bias in estimating individual age, it should affect age for both sexes in the same way, in which case SAD should remove any such bias. In our study, the potential sources of error resulting from natural and experimental constraints are likely to constitute only random noise. Therefore, the error should not affect systematically the results.

In conclusion, our results demonstrate hyperallometry in SSD in a species where the sexes are that females being larger than males. This translates to a pattern following an inverse of Rensch's rule. Fecundity selection plays the important role in the inverse Rensch's rule because large females increase reproductive output than small males within and among populations. The variation of SSD in the species can be explained in terms of differences in age between the sexes for populations.

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References

- Abouheif E, Fairbairn DJ (1997) A comparative analysis of allometry for sexual size dimorphism: assessing Rensch's rule. *Am Nat* 149:540–562
- Andersson M (1994) Sexual selection. Princeton University Press, Princeton
- Blanckenhorn WU (2005) Behavioural causes and consequences of sexual size dimorphism. *Ethology* 111:977–1016
- Blanckenhorn WU, Dixon AFG, Fairbairn DJ, Foellmer MW, Gibert P, van der Linde K, Meier R, Nylin S, Pitnick S, Schoff C, Signorelli M, Teder T, Wiklund C (2007) Proximate causes of Rensch's rule: does sexual size dimorphism in arthropods result from sex differences in developmental time? *Am Nat* 169:245–257
- Chen BY, Liao WB, Mi ZP (2011a) Body size and age of the China Wood Frog (*Rana chensinensis*) in northeastern China. *North-West J Zool* 7:236–242
- Chen W, Zhang LX, Lu X (2011b) Higher Pre-hibernation energy storage in anurans from cold environments: a case study on a temperate frog *Rana chensinensis* along a broad latitudinal and altitudinal gradients. *Ann Zool Fennici* 48:214–220
- Colwell RK (2000) Rensch's rule crosses the line: convergent allometry of sexual size dimorphism in hummingbirds and flower mites. *Am Nat* 156:495–510
- Dale J, Dunn PO, Figuerola J, Lislevand T, Székely T, Whittingham LA (2007) Sexual selection explains Rensch's rule of allometry for sexual size dimorphism. *Proc R Soc Lond B* 274:2971–2979
- Fairbairn DJ (1997) Allometry for sexual size dimorphism: pattern and process in the coevolution of body size in males and females. *Ann Rev Ecol Syst* 28:659–687
- Fairbairn DJ, Blanckenhorn WU, Székely T (2007) Sex, size, and gender roles: evolutionary studies of sexual size dimorphism. Oxford University Press, Oxford
- Hemelaar ASM (1981) Age distribution of male *Bufo bufo* (Amphibia: Anura) from the Netherlands, based on year rings in phalanges. *Amphib-Reptilia* 1:223–233
- Herczeg G, Gonda A, Merilä J (2010) Rensch's rule inverted—female-driven gigantism in nine-spined stickleback *Pungitius pungitius*. *J Anim Ecol* 79:581–588
- Lengkeek W, Didderen K, Cote IM, van der Zee EM, Snoek RC, Reynolds JD (2008) Plasticity in sexual size dimorphism and Rensch's rule in mediterranean blennies (Blenniidae). *Can J Zool* 86:1173–1178
- Liao WB (2011) A skeletochronological estimate of age in a population of the Siberian Wood Frog, *Rana amurensis*, from northeastern China. *Acta Herpetol* 6:237–245
- Liao WB, Lu X (2010a) Age structure and body size of the Chuanxi tree frog *Hyla annectans chuanxiensis* from two different elevations in Sichuan (China). *Zool Anz* 248:255–263
- Liao WB, Lu X (2010b) A skeletochronological estimation of age and body size by the Sichuan torrent frog (*Amolops mantzorum*) between two populations at different altitudes. *Anim Biol* 60:479–489
- Liao WB, Lu X (2011) Variation in body size, age and growth in the Omei Treefrog (*Rhacophorus omeimontis*) along an altitudinal gradient in western China. *Ethol Ecol Evol* 23:248–261
- Liao WB, Lu X (2012) Adult body size= f (initial size+growth rate \times age): explaining the proximate cause of Bergman's cline in a toad along altitudinal gradients. *Evol Ecol* 26:579–590
- Liao WB, Zhou CQ, Yang ZS, Hu JC, Lu X (2010) Age, size and growth in two populations of the dark-spotted frog *Rana nigromaculata* at different altitudes in southwestern China. *Herpetol J* 20:77–86
- Liao WB, Lu X, Shen YW, Hu JC (2011) Age structure and body size of two populations of the rice frog *Rana limnocharis* from different altitudes. *Ital J Zool* 78:215–228
- Lu X (1994) Feature of fecundity and its geographic variation in *Rana chensinensis*. *Acta Ecol Sin* 14:209–214
- Lu X, Li B, Liang JJ (2006) Comparative demography of a temperate anuran, *Rana chensinensis*, along a relatively fine altitudinal gradient. *Can J Zool* 84:1789–1795
- Ma XY, Tong LN, Lu X (2009) Variation of body size, age structure and growth of a temperate frog, *Rana chensinensis*, over an elevational gradient in northern China. *Amphibia-Reptilia* 30:111–117
- Monnet JM, Cherry MI (2002) Sexual size dimorphism in anurans. *Proc R Soc Lond B* 269:2301–2307
- Polák J, Frynta D (2009) Sexual size dimorphism in domestic goats, sheep, and their wild relatives. *Biol J Linn Soc* 98:872–883
- Polák J, Frynta D (2010) Patterns of sexual size dimorphism in cattle breeds support Rensch's rule. *Evol Ecol* 24:1255–1266
- Remeš V, Székely T (2010) Domestic chickens defy Rensch's rule: sexual size dimorphism in chicken breeds. *J Evol Biol* 23:2754–2759
- Shine R (1979) Sexual selection and sexual dimorphism in the Amphibia. *Copeia* 1979:297–306
- Sokal RR, Rohlf FJ (1981) Biometry: the principles and practice of statistics in biological research. Freeman, WH, San Francisco
- Székely T, Freckleton RB, Reynolds JD (2004) Sexual selection explains Rensch's rule of size dimorphism in shorebirds. *PNAS* 101:12224–12227
- Xie F, Ye CY, Fei L, Jiang JP, Zeng XM, Matsu M (2000) Taxonomical studies on *Rana chensinensis* in northwestern China (Amphibia: Ranidae). *Acta Zootaxon Sin* 25:228–235