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## Oviposition site selection by rice frogs on Taohua Island and the nearby mainland

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Oviposition site selection influences adult reproductive success as well as offspring fitness. Between 2007 and 2009 we studied oviposition site selection of the rice frog (*Fejervarya limnocharis*), one of the most abundant frogs in China, on Taohua Island and the nearby mainland in eastern China. The distance to the nearest road affected oviposition site selection both on Taohua island and the mainland. On the mainland, vegetation coverage also has positive effects on oviposition site selection. We suggest that human disturbance and predator pressure affect rice frog oviposition site selection at both sites.

**Key words:** distance to the nearest road, human impact, predator pressure, rice frog, vegetation coverage

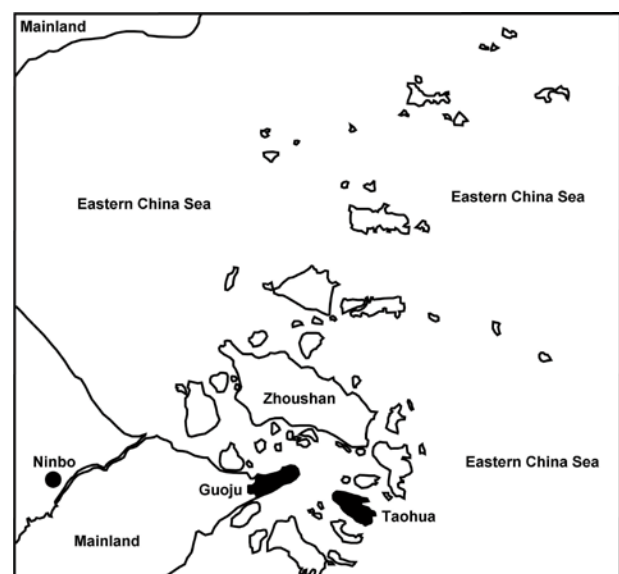
In amphibians, oviposition site selection influences adult reproductive success as well as the fitness of the resulting offspring, by directly or indirectly affecting hatching success and larval growth, development and survival (Resetarits & Wilbur, 1989). Poor oviposition site choice can result in increased predation on offspring (Sargent & Gebler, 1980; Hatchwell et al., 1996; Downes & Shine, 1999; Kolbe & Janzen, 2001), reduced hatching success (Cagle et al., 1993; Wilson, 1998; Warner & Andrews, 2002) and reduced offspring fitness (Shine & Brown, 2002). Finding a suitable oviposition site is especially important for species with no maternal care after oviposition (Kolbe & Janzen, 2001; Blouin-Demers et al., 2004; Hughes & Brooks, 2006).

The rice frog (*Fejervarya limnocharis*) is one of the most common amphibians in China, occurring in rice fields, rivers, ponds and reservoirs (Huang et al., 1990; Fei et al., 1999). The goal of the present paper is to characterize and compare oviposition sites of rice frogs on Taohua Island in the Eastern China Sea and the

nearby mainland. Both sites are characterized by similar environmental and climatic conditions, and abundant occurrences of rice frogs (Gu & Jin, 1985; Chen, 1989; Wu et al., 2006).

Taohua Island (about 41 km<sup>2</sup> in size; part of the Zhoushan Archipelago) in the Eastern China Sea was separated from mainland of Guoju in the northeastern part of Zhejiang province about 7000–9000 years ago, due to rising sea levels during the late Pleistocene (Fig. 1; Jinling, 1987; Chen, 1989; Wang et al., 2009). Both the island and the nearby mainland have a climate typical for the subtropical ocean monsoon zone, and are covered with subtropical evergreen broadleaf forests (Huang et al., 1990; Zhuge & Gu, 1990). The mean temperature ranges from 5.7°C in January to 26.7°C in July on Taohua Island, and from 5.2°C (January) to 27.8°C (July) on the mainland (Gu & Jin, 1985; Huang et al., 1990; Zhuge & Gu, 1990). Ten amphibian species are found on Taohua Island, and 17 species are on the mainland (Huang et al., 1990; Yiming et al., 1998; Fei et al., 1999). On both sites, rice frogs generally emerge from hibernation in March and breed between early April and early September (Huang et al., 1990). They lay eggs in various lentic water or water-filled depressions such as rice fields, ditches, marshes and small pools (Huang et al., 1990; Wu et al., 2006).

Our study was conducted from May to August during 2007–2009. We recorded rice frog oviposition habitats on rainless days between 0500 and 1600 hours, by carefully searching each potential breeding site for egg clutches.



**Fig. 1.** Map of sample sites of the Taohua Island and Guoju on the adjacent mainland in eastern China.

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**Table 1.** Comparison of habitat variables between oviposition sites and control sites on the Taohua Island and the nearby mainland (significant results in *italics*).

	Variables	Oviposition site (Mean±SE)	Control site (Mean±SE)
Mainland	Water (%)	45.04±1.47	46.54±1.40
	Vegetation cover (%)	<i>30.51±1.54</i>	<i>26.12±1.51</i>
	Water depth (cm)	8.45±0.67	7.42±0.41
	pH	7.03±0.03	7.04±0.03
	Distance to the nearest road (m)	<i>11.39±0.44</i>	<i>6.36±0.50</i>
Taohua island	Water (%)	42.34±1.45	43.50±1.14
	Vegetation cover (%)	32.97±1.52	32.59±1.57
	Water depth (cm)	8.42±0.38	8.92±0.82
	pH	7.03±0.02	7.03±0.02
	Distance to the nearest road (m)	<i>9.85±0.53</i>	<i>4.22±0.41</i>

Once an egg clutch was found, we placed a one-metre square grid over the centred egg clutch, and recorded water pH, distance to the nearest road, percentage of water coverage, vegetation coverage and water depth within the grid. The one-metre square grid was divided into 100 sub-grids, enabling to estimate water coverage and vegetation coverage with an accuracy of 1%. Control sites were selected by choosing two random numbers indicating direction (0 to 360 degrees) and distance from the actual oviposition site (0–50 m). The same habitat variables as taken at the oviposition sites were recorded at the control sites (Table 1).

In 2007, 2008 and 2009 we found 17, 26 and 24 oviposition sites on the mainland, and 21, 23 and 24 oviposition sites on the Taohua Island, respectively. There was no significant difference for all habitat features during the three years of our study in either the mainland population (water coverage:  $\chi^2=1.807$ ,  $df=2$ ,  $p=0.405$ ; vegetation coverage:  $\chi^2=0.677$ ,  $df=2$ ,  $p=0.713$ ; water depth:  $\chi^2=0.982$ ,  $df=2$ ,  $p=0.612$ ; water temperature:  $\chi^2=0.896$ ,  $df=2$ ,  $p=0.639$ ; distance to the nearest road:  $\chi^2=0.272$ ,  $df=2$ ,  $p=0.873$ ) or the Taohua Island population (water coverage:  $\chi^2=0.742$ ,  $df=2$ ,  $p=0.690$ ; vegetation coverage:  $\chi^2=0.265$ ,  $df=2$ ,  $p=0.876$ ; water depth:  $\chi^2=2.161$ ,  $df=2$ ,  $p=0.339$ ; water temperature:  $\chi^2=0.562$ ,  $df=2$ ,  $p=0.755$ ; distance to the nearest road:  $\chi^2=0.565$ ,  $df=2$ ,  $p=0.754$ ). We therefore pooled the three years to enlarge the sample size.

A Kolmogorov-Smirnov test revealed that all habitat variables were not normally distributed (detailed data

not shown), and we used Kruskal-Wallis tests to detect the differences in the habitat variables between the oviposition sites and control sites on the island and the mainland (Zar, 1999). We performed the statistical analyses using SPSS v17.0 (2008) and determined statistical significance at the level of  $p>0.05$ . We used generalized linear models to detect the determinants of oviposition site selection on the island and the nearby mainland. We determined a minimum adequate model using Akaike's Information Criterion (AIC) with the lowest AIC value after fitting the full model (in total we ran 120 models, Burnham & Anderson, 2002; Richards, 2005). The analyses were conducted with R version 2.8.1 (R Core Development Team, 2006).

On Taohua Island, the distance to the nearest road was the only variable that differed between oviposition sites and control sites (Kruskal-Wallis tests,  $\chi^2=53.041$ ,  $df=1$ ,  $p<0.001$ ). On the nearby mainland, vegetation coverage and distance to the nearest road were different between oviposition sites and control sites (Kruskal-Wallis tests,  $\chi^2=10.379$ ,  $df=1$ ,  $p=0.001$  for vegetation coverage;  $\chi^2=41.374$ ,  $df=1$ ,  $p<0.001$  for distance to the nearest road, Table 1). Distance to the nearest road influenced rice frog oviposition site selection on both Taohua Island and the mainland, and vegetation coverage influenced rice frog oviposition site selection on the mainland (Table 2).

Distance to the nearest road affected both the island and the mainland oviposition sites. Roads generally negatively impact behavioural variables of wildlife (Wang et al., 2008). Roads are also used by small mammals,

**Table 2.** Results of the minimum adequate model based on generalized linear models with the rice frog oviposition sites to the explanatory variables on the Taohua Island ( $n=68$ ) and nearby mainland ( $n=67$ ). Non-significant variables are not reported. The multivariate model was the best model according to the Akaike's Information Criterion (AIC).  $\beta$ : regression coefficient;  $p$ : likelihood Ratio Test. The AIC for the minimum adequate model was 124.39 for the Taohua Island and 136.01 for the nearby mainland.

Location	Explanatory variables	$\beta$	SE	$p$
Taohua island	Distance to the nearest road	7.48	1.33	<0.001
Mainland	Vegetation coverage	1.79	1.03	0.039
	Distance to the nearest road	7.50	1.45	<0.001

snakes, pet dogs and cats, which can be predators of amphibians (Li et al., 2011). In this study, distance to the nearest road influenced oviposition sites at both sites, suggesting that predators along roads may be responsible for oviposition site selection in rice frogs.

Vegetation coverage had an effect on oviposition site selection on the mainland, but not on Taohua Island, which might reflect differential predatory pressures at these sites. Pressure from predation has been generally known to be weaker on islands than the mainland (Adler & Levins, 1994), and has also been shown for Taohua Island (Li et al., 2011). Vegetation serves as a habitat for predator avoidance on both Taohua Island and on the mainland, and its uneven use might reflect the predator pressure difference at these two sites.

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