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Spatial variation in breeding phenology at small spatial scales: a stochastic effect of population size

Abstract

Spatial variation in phenology can occur at small spatial scales over which individuals can disperse or forage within one generation. Previous studies have assumed that variations in phenological peaks are caused by differences in abiotic environmental characteristics. However, environments should generally be similar among local habitats over small spatial scales. When the local population size is small, the phenological peak of the local population should be strongly affected by the variation in timing expressed by individuals. If a regional population consists of small local subpopulations (e.g. a metapopulation), the stochastic processes regulated by population sizes may explain the spatial variation in phenology. In this study, we quantitatively evaluated the extent of the spatial and annual variations in the breeding phenology of the forest green tree frog, *Rhacophorus arboreus* habiting a small area (< 10 km^2). The spatial variation in phenological peaks among 25 breeding sites was large over six years. This spatial variation was not explained by differences in air temperature or water depth. Randomization tests revealed that a large portion of the spatial variation could be explained by differences in population size, without considering site-specific factors. Annual variations in phenological peaks tended to be greater for smaller populations. These results imply that the stochastic process might have caused the spatial and annual variations in the phenological peaks of *R. arboreus* observed in the study region. Understanding spatiotemporal variation in phenology determined by stochastic process would be important to better predict interspecific interactions and (meta)population dynamics at small spatial scales.

Keyword

Amphibian; Demographic stochasticity; Metapopulation; Phenology; Spatial variation

Introduction

Phenological variations among populations over small spatial scales, such as those over which individuals can forage or disperse within a single generation, can have unique consequences for interspecific interactions and population dynamics at local (i.e., within a certain population) and regional (i.e., when the population consist of several local subpopulations in a certain region) scales. For example, when mature individuals have to disperse across local habitats to find their mates, spatial variation in reproductive phenology at the regional scale can decrease the reproductive success of dispersers (i.e., due to reproductive asynchrony; Walter et al. 2015). On the other hand, consumers can extend their foraging opportunities by tracking the spatial variation in the phenology of their food resource at the regional scale when the resource availability in each local habitat is temporally limited (i.e., phenological tracking; Post et al. 2008; Deacy et al. 2016). These ecological consequences will depend on the degree and patterns of spatial variations in phenology (Armstrong et al. 2016; Takahashi and Sato 2017). However, few studies have quantified spatial variation in phenology, and the factors causing phenological variations at small spatial scales are still poorly understood.

Previous studies have implicitly assumed that environmental factors cause variations in phenology (Fig. 1a). For example, variation in water temperature among tributaries influences the phenology of the peak breeding periods of sockeye salmon populations (Ruff et al. 2011). However, the differences in environmental characteristics among habitats should generally decrease as the spatial scale considered decreases (Koenig 1999), which allows us to infer that other factors may explain the phenological variations that occur over small spatial scales. Specifically, when the local population size is very small, then the phenological peak of the local population should be strongly affected by the variation in timing expressed by individuals (Fig. 1b). If a regional population consists of small local subpopulations (e.g., a metapopulation; Hanski 1998), the unique stochastic processes occurring therein may explain the spatial variation in phenology over small spatial scales; herein, we define this process as ‘demographic stochasticity in phenology’.

Demographic stochasticity in phenology could also explain the magnitude and direction of annual shifts in the spatial variations in phenology, which may be important for stabilizing or destabilizing population dynamics (Post et al. 2008). For instance, the phenology of a small population would shift annually without a particular directionality as a result of this demographic stochasticity (e.g., the phenological peak can be early in one year, but can turn out to be late in another year in the same local population), while that of a large population should be relatively constant (Fig. 1b). This would further drive the annual variation in the spatial variations in phenology. On the other hand, if the spatial variation in phenology is driven by environmental factors, the annual shifts in the phenology of local populations would show a similar trend among local populations in the same region. As a result, the annual variation in the spatial variation among populations might be small (but see Post 2003).

The forest green tree frog, *Rhacophorus arboreus* (Anura: Rhacophoridae) is an endemic amphibian species in Japan. Previous studies have shown that *R. arboreus* generally has a prolonged breeding period, which lasts from April to July depending on the breeding region considered (Kato 1955). During this species' annual breeding season, a female spawns one foam nest on a tree branch or on the land, which is located nearby water (Fig. S1). Therefore, the number of foam nests in each site indicates the population size of breeding females in each site. In the region of the present study, we reported that the total number of foam nests of *R. arboreus* varied from 3 to 104 among 14 breeding sites (Takahashi and Sato 2015), and the spatial variation in the phenology of the peak breeding period was also substantial (a maximum variation of 17 days), all within a relatively small forested area (< 10 km^2) in a single-year survey. This spatial variation is expected to greatly affect the fitness of *R. arboreus* because it allows a predator, the Japanese fire-bellied newt (*Cynops pyrrhogaster*), to undergo phenological tracking, which can decrease the survival rates of *R. arboreus* tadpoles (Maeda and Matsui 1999; Takahashi and Sato 2017). Altitude could not explain the spatial variation observed across this small region, and other environmental factors that are important to amphibian breeding phenology, such as air temperature and water depth, seemed to be similar among local breeding sites (Takahashi and Sato 2015). Furthermore, the regional population consists of more than 10 small local subpopulations (plus a few large subpopulations), which allows us to infer that there is likely demographic stochasticity in phenology within this system.

The purpose of this study was to quantify the spatial and annual variations in phenology over a small spatial scale, and to examine the potential effect of population size on phenological variations occurring among populations. To this end, we examined the phenology of the peak breeding period of *R. arboreus* in 25 breeding sites across six years, and tested whether: (1) the phenology of the peak breeding period differed significantly among sites, years, or both; (2) the observed spatial variation was explained by population size or a combination of population size and spatial environmental variation and (3) the annual variation in each site was larger with smaller population size. After running our analyses, the results suggested that the demographic stochasticity in phenology could be important in explaining the observed spatial and annual variations in the breeding phenology of *R. arboreus* among local populations in the study region.

Material and methods

Field observations

Field observations occurred from May to August from 2014 to 2019 in the Ashiu Forest Research Station (AFRS), Kyoto University, Kyoto, Japan (35°19′N, 135°43′E). In the AFRS, *R. arboreus* is a common amphibian species. Over these six years, we identified 25 breeding sites, which were small temporary ponds and marshes located along streams (Table 1; Fig. 2). We treated these breeding sites as local populations. Excluding 2014, newly spawned foam nests were recorded in each site every 1–3 days, with these observations covering most of the breeding season in each year. Since larvae usually hatch in a foam nest and then drop into ponds approximately two weeks after being laid, there is a risk that such disturbances as wind, heavy rain, and predation by newts can result in the collapse of foam nests before two weeks elapse, potentially leading to underestimated numbers of nests if they are not observed frequently enough; therefore, our sampling interval was designed to avoid underestimating the number of foam nests in the event of such disturbances. However, in 2014, field surveys were conducted every 7–15 days.

The area of each site was measured in the breeding seasons in 2017 and 2018. Air temperature in each site was measured every one hour using thermometers (KN Laboratories, Osaka, Japan) from May to September in 2018. Precipitation is an essential factor for amphibian breeding phenology. In AFRS, the mean annual precipitation was 2,257 mm (average from 1981 to 2010; FSERC 2003), and the rainy season lasts from May to July. Because of the high precipitation during the breeding season, we assume that there is relatively little difference in precipitation among sites across the small spatial scale. However, precipitation might influence breeding phenology indirectly through water depth. In AFRS, some ponds are temporal, and *R. arboreus* start to breed after the appearance of the ponds (Takahashi, personal observation). Therefore, we measured water depth at one to six locations in each site every three days in 2018.

Data analysis

All statistical analyses were conducted in R 3.5.1. To test the extent of the spatial and annual variations in the breeding phenology of *R. arboreus*, we used an accelerated failure time (AFT) model with a log-normal distribution (Kalbfleisch and Prentice 2002), which is a parametric survival analysis model. Generally, the survival analysis estimates the time until some event occurs (e.g., death). We used the AFT model to estimate the time until an individual breeding event occurs. Therefore, we used a day of the year when the foam nest was observed for the first time as a response variable. Since we used the survey days conducted with 1–15 interval days as dates of the breeding events, we might overestimate the actual breeding date especially in 2014 because of long intervals. However, we believe that it might not affect the relative difference in phenology among sites because the above situation should be same among sites. Breeding sites, years, and their interaction were used as explanatory variables. Model selection was conducted using Akaike's information criterion (AIC) to test whether phenological variation was better explained by differences among the breeding sites, years, and/or their interaction.

The relationships between the phenological peaks in each population and the environmental factors in each site were analyzed using a generalized linear model (GLM) with a Gaussian distribution. To quantify spatial variation in phenology, the median breeding date was calculated to represent the phenological peak in each year at each local site (i.e., for each local population). We discovered some breeding sites near the end of the breeding period. Hence, in all our analysis using the median date, we only included breeding sites present on at least 90 % of our observation days in each year. In this model, the median breeding date of each population in 2018 was used as a response variable, while the mean air temperature in June, mean water depth in June, and their interaction were included as explanatory variables. Model selection was then conducted using AIC.

We performed randomization tests to assess whether the spatial variation in phenological peaks in each year could be explained by demographic stochasticity.

Randomization tests are useful to compare observed data and the expected distribution of the data due to some random process while taking the population size into account (Caswell 2001; Fréville et al. 2004). The principle of this test is to assess whether site-specific factors (such as environmental ones) should cause the observed phenological peaks in each population to occur earlier or later than those predicted by a random (stochastic) process that depends on population size. Thus, the null hypothesis is represented by a distribution of the timing of individual breeding events that is derived from demographic stochasticity but is independent of site-specific factors. This randomization test cannot directly test the effects of demographic stochasticity. However, failing to reject the null hypothesis in this test implies that the phenological peaks in each population can be explained by demographic stochasticity alone, without considering the effects of site-specific factors (see Discussion). In the randomization test, we calculated the phenological peak of each population in each year as the median breeding date, as described above. To estimate the distribution of the median breeding date under the null hypothesis, we generated a simulated phenology in each population by randomly permuting individual breeding dates from the regional population (i.e., all individuals from the whole study site) in a certain year. In the permutation process, we maintained the observed total number of foam nests in each population (i.e., population size) and the total number of foam nests seen on each day of observations in the regional population (i.e., the whole study area). In doing so, the simulated median breeding date for each population was obtained in each permutation, while maintaining the natural structure of the local populations and regional-scale phenology of the study site. We iterated the simulation 10000 times to obtain the distribution of the median breeding date under the null hypothesis. Then, a two-tailed test was conducted for each population and each year to determine whether the observed median date was outside of the estimated randomized distribution. We repeated randomization tests 98 times, which might cause type I error in some site-year combinations and overestimate the number of significant site-year combinations, which lead to overestimate the influence of the environmental factors in determining phenology. However, if we adjust the p-value for the multiple comparisons, it might also cause type II error. In this study, we are interested not only in the alternative hypothesis but also in the null hypothesis. In this sense, Type II error induces overestimation of the influence of demographic stochasticity, which should also be minimized because our claim is the importance of the demographic stochasticity. Therefore, we did not correct multiple comparisons and used a conservative significant level (α = 0.05).

In addition, we tested to what extent the observed spatial variation at the regional scale in each year could be explained by the effect of demographic stochasticity. To this end, we calculated the coefficient of variation (CV = standard deviation/mean) of the phenological peaks across all sites as a statistic representing the spatial variation for each of the 10000 permutations performed. A one-tailed test (α = 0.05) was then used to assess whether the significance of the observed CVs of spatial variation in each year was greater than the estimated randomized distribution of CVs. As described above, we did not correct multiple comparisons to avoid type II error.

To evaluate the extent of the annual variation in phenological peaks in each site, the CV of the median breeding date was calculated. The relationship between the CV and the mean population size across six years observed in each site was tested using Pearson's correlation tests (α = 0.05).

Results

In the AFRS, the breeding period of *R. arboreus* extended from May 12 (in 2015) to August 13 (in 2015). We found 0–42 foam nests each site each day. The total annual number of foam nests seen in the AFRS per year ranged from 322 (in 2014) to 614 (in 2019). The low number of nests in 2014 could be due to environmental conditions or because we surveyed for nests less frequently than other years. The total annual number of foam nests observed in each site ranged from 5–103 (in 2015) to 2–219 (in 2018). The total annual number of foam nests in each site was highly correlated between consecutive years (Pearson's correlation, r = 0.87–0.98).

The AFT model revealed that the days of the year when the foam nests were observed were best explained by the additive effects of sites and years. The interaction term was not included in the best model selected by AIC (Table S1; ΔAIC = 32.6).

The median breeding dates in the AFRS region ranged from June 11 (in 2016) to June 26 (in 2019). The maximum difference in the median dates among sites ranged from 15.5 days in 2014 to 33.5 days in 2018 (Fig. 3). Neither the air temperature nor the water depth in June 2018 were significantly related to the median breeding dates seen in 2018, as no GLM containing such fixed terms was selected as the best model during the AIC model selection procedure (ΔAIC = 4.7 between the null model and the full model).

In randomization tests, when comparing observed median breeding dates to null median breeding dates, we found that more than half of the site-year combinations were not significantly different from the null (Table S2). Thus, much of the observed phenological variation in median breeding date was not necessarily explained by site-specific factors in each population. For the 31 out of 98 site-year combinations across the six years where the null hypothesis was rejected, the observed median dates were significantly later than those generated under the null hypothesis in 25 site-year combinations, while the peaks were earlier in six other site-year combinations.

When comparing observed CVs of breeding dates to null CVs, the null hypothesis was not rejected in four of the six years (2015 and 2016 were the exceptions; Fig. S2), which meant that site-specific factors were not necessarily needed to explain the spatial variation in median breeding date in AFRS.

The maximum interannual variation in the median breeding dates in each site across the six years ranged from 2.5 days in site 12 to 23.0 days in site 5. The CVs of the smaller populations tended to be larger than those of the larger populations, although the relationship between population size and CV was not statistically significant (Fig. 5; Pearson's correlation test, $r = -0.35$, $p = 0.12$).

Discussion

Although many studies have demonstrated spatial variation in phenology at relatively large spatial scales (Sheridan et al. 2018), it has rarely been quantified at small spatial scales, despite its potentially important roles in mediating interspecific interactions and population dynamics (Post et al. 2008). In this study, significant spatial and annual variations in the breeding phenology of *R. arboreus* were observed over a relatively small spatial scale (< 10 km^2) across six years. Moreover, the results of our analyses suggested that demographic stochasticity could partially explain the observed spatiotemporal variation in phenology. Previous studies of conservation biology and population genetics have demonstrated the role of stochasticity in population dynamics (Bartlett 1960; Fréville et al. 2004). However, to the best of our knowledge, this is the first study inferring that demographic stochasticity is a factor mediating spatial variation in phenology.

Although the underlying mechanisms of the demographic stochasticity observed in this study, such as factors causing variations in the phenology of individuals, are still unknown, these results confirm that describing variations in phenology at the population level based on demographic stochasticity should be useful for predicting interspecific interactions and (meta)population dynamics at small spatial scales (Forrest and Miller-Rushing 2010; Inouye et al. 2019).

Phenological asynchrony among habitats is thought to increase predation pressure from mobile predators (Ims 1990). In the study area (< 10 km^2), large spatial variations in phenological peaks (more than 2 weeks) were observed in every year. This spatial variation in breeding phenology is critical for the survival of *R. arboreus* tadpoles in spatially structured habitats (Takahashi and Sato 2017). When there is a considerable spatial variation in phenology among local populations, phenological tracking by newt predators can more strongly reduce the survival rates of local populations of *R. arboreus* than when there is synchronized phenology among populations.

Air temperature and water depth are important environmental factors affecting amphibian breeding phenology (Lane and Mahony 2002; Sheridan et al. 2018). However, even in 2018, when large spatial variation was observed in these factors, they still did not explain the spatial variation in the phenological peaks in our study region very well. Results of the randomization tests done at the regional scale implied that the spatial variation in phenology (at least in 2014, 2017, 2018 and 2019) might be explained by differences in population size without needing to refer to any site-specific factors, such as the environmental characteristics of each site. However, this does not mean that the environmental factors did not affect breeding phenology in these years. In fact, results of the randomization tests at the local scale suggested that, to explain the phenological peak in some sites (sites 20, 21, 22, and 23), it was necessary to take the effects of site-specific factors into consideration. These sites are located in an area of the AFRS that is dried marshland, at least early in the breeding season, but became a wet marsh later in the breeding season (Table 1). As the phenological peaks of these sites were often significantly later than those predicted by the null hypothesis (Table S2), the delayed increase in water depth may be a cue for *R. arboreus* to begin breeding in these study sites.

Interannual variation in peak breeding phenology varied from 2.5 days to 23.0

days, and tended to increase with decreasing population size, which is another piece of circumstantial evidence for the importance of demographic stochasticity. Because small populations are sometimes important in metapopulation dynamics (Hanski et al. 1995), differences in interannual variation caused by demographic stochasticity and environmental factors might lead to different predictions of regional population dynamics (or metapopulation dynamics). For example, spatial heterogeneity in environments can sustain amphibian metapopulations at small spatial scales (McCaffery et al. 2014; Heard et al. 2015). Although a simple correlation test did not reveal a significant relationship between the phenological peaks and local habitat characteristics observed in the present study, the relative importance of demographic stochasticity varied among years (Fig. S2; Table S2). For example, spatial variation in phenology might be low in low-precipitation years because breeding opportunities in them might be limited. In such years, it would be difficult to detect effects of demographic stochasticity because this environmental factor would reduce the possible variations in individual phenology at the regional scale. Future studies will be necessary to evaluate the relative importance of demographic stochasticity and environmental factors in determining spatial variation in phenology so that population dynamics can be better predicted at small spatial scales.

A metapopulation often consists of small subpopulations (Hanski et al. 1995). Spatial and temporal variations in phenology may be more commonly caused by demographic stochasticity in such a system. However, there are some situations wherein the importance of demographic stochasticity to the spatial variation in phenology would be limited. For example, demographic stochasticity might not cause large spatial variations in the phenology of explosive breeders, which start to reproduce rapidly and at high levels after they receive particular environmental cues. This is because breeding in species with such reproductive systems is highly synchronized by environmental cues, and hence the potential variation in individual phenology would be low. Moreover, if genetic variation for phenological traits is limited in a regional population, it would be difficult for demographic stochasticity in phenology to occur. Future studies should determine in which situations demographic stochasticity will be an important factor causing ecologically important variations in phenology.

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Figure and Table legends

Table 1. Physical characteristics of the breeding sites observed in the Ashiu Forest Research Station (AFRS), Kyoto, Japan. Mean air temperatures in June 2018 and mean water depths in June 2018 or 2019 in each site are also shown. Some sites could not observed in 2018 and 2019.

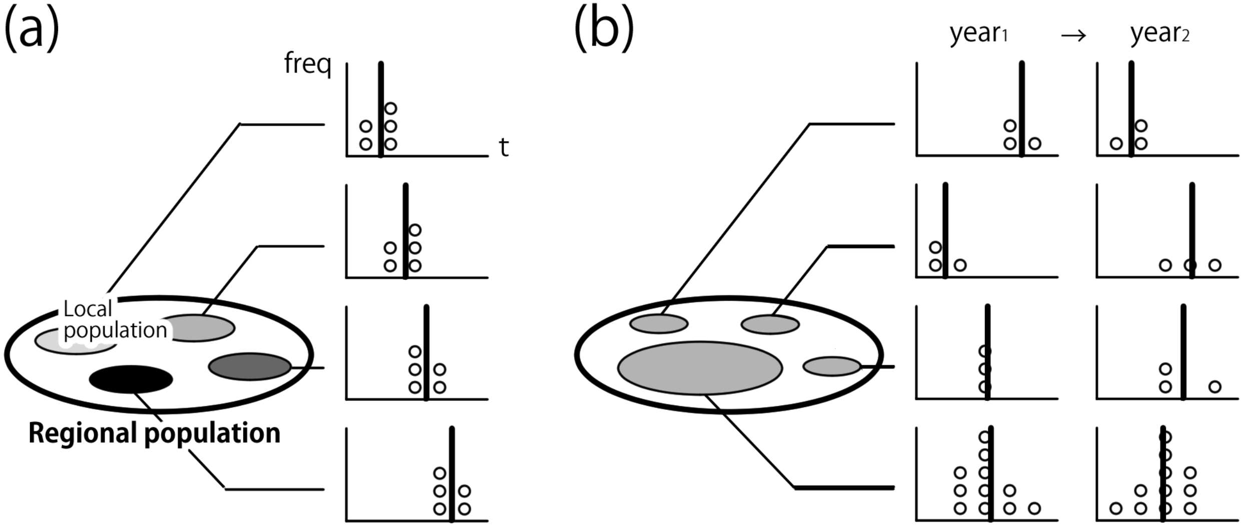
Figure 1. Schematic diagram of spatial variations in phenological peaks among local populations caused by (a) site-specific environmental effects and (b) demographic stochasticity in phenology. In each figure, the outer circle represents the regional population, while the inner circles represent local subpopulations. The colors of the inner circles denote an environmental gradient that influences phenology. The sizes of the inner circles denote the sizes of local subpopulations. The right-hand graphs show the phenology of the local subpopulations. Each axis shows time (date) and frequency. Solid lines and dots on the graphs denote the phenological peaks of local subpopulations and individual phenology, respectively.

Figure 2. Locations of the 25 breeding sites in the AFRS. Raw data for this location map were obtained from the electronic contour map 25000 published by the Geospatial Information Authority of Japan.

Figure 3. Cumulative proportions of the total numbers of foam nests observed in each site from June 1 to July 16 in (a) 2014, (b) 2015, (c) 2016, (d) 2017, (e) 2018, and (f) 2019. Numbers in parentheses represent the total number of foam nests observed in each site. Line colors correspond to those of the closed (inner) circles in Figure 1. Site 3 dried up from 2015 to 2018.

Figure 4. Relationship between the mean number of foam nests observed across six years and the annual variation in peaks of breeding phenology. Line colors correspond to the sites shown in Figure 1.

Sites	Area (m^2)	Temperature (°C)	Depth (cm)
1	3.68	17.73	17.8
2	17.02	17.91	36.8
3	-	-	-
4	2.33	17.49	28.2
5	11.40	17.02	4.1
6	3.87	17.22	1.5
7	2.08	16.32	5.6
8	7.03	-	1.2
9	6.68	16.77	25.9
10	2.38	15.94	16.8
11	2.61	16.26	3.1
12	7.83	16.54	19.1
13	3.21	16.12	8.9
14	7.90	16.48	7.7
15	-	-	-
16	1.87	15.24	5.9
17	24.38	16.19	2
18	48.57	16.38	2.7
19	61.36	-	3
20	74.59	17.28	13.5
21	-	-	-
22	25.58	17.02	1.4
23	0.85	16.82	0.4
24	51.08	17.18	3.4
25	422.96	17.32	4.1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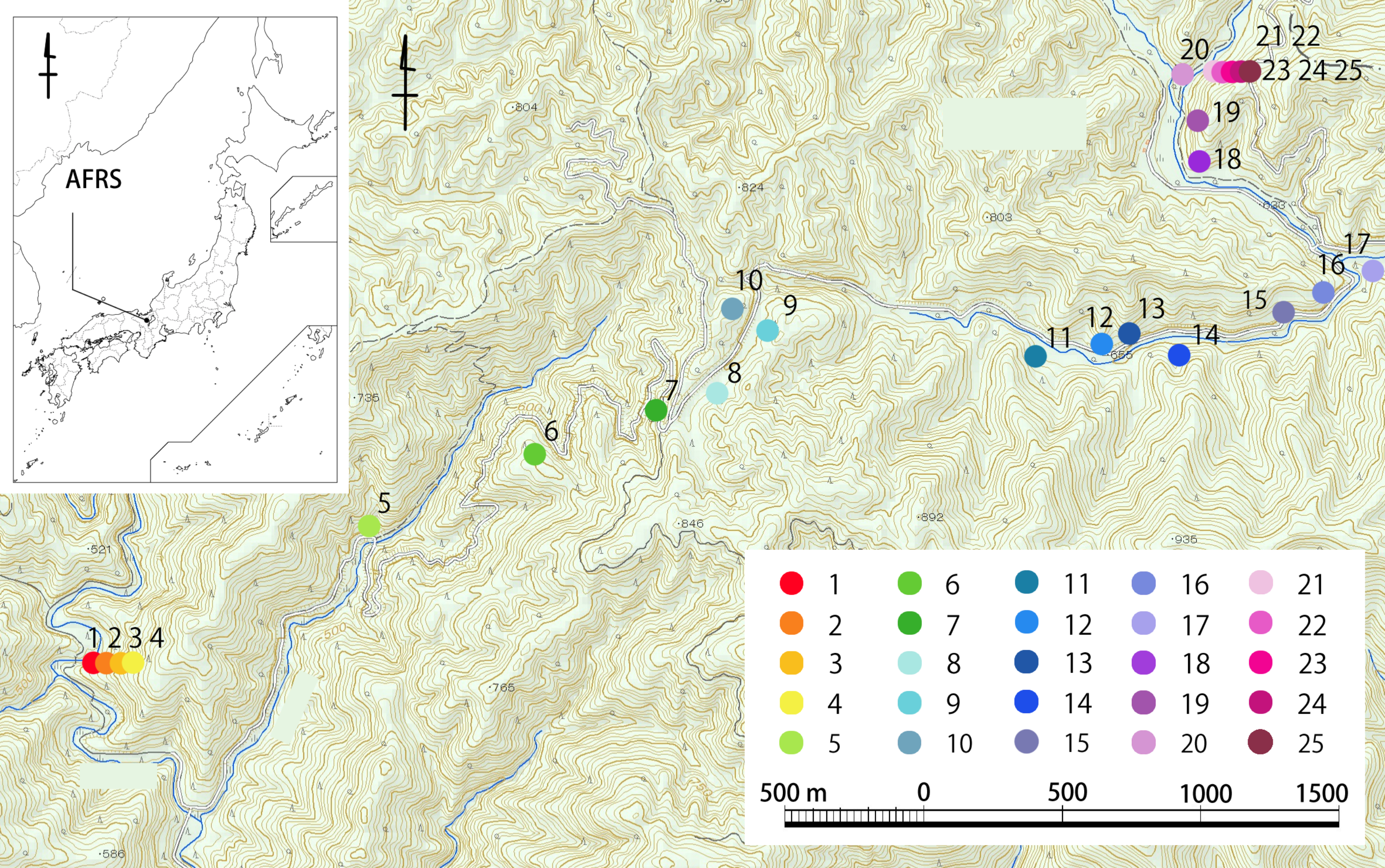
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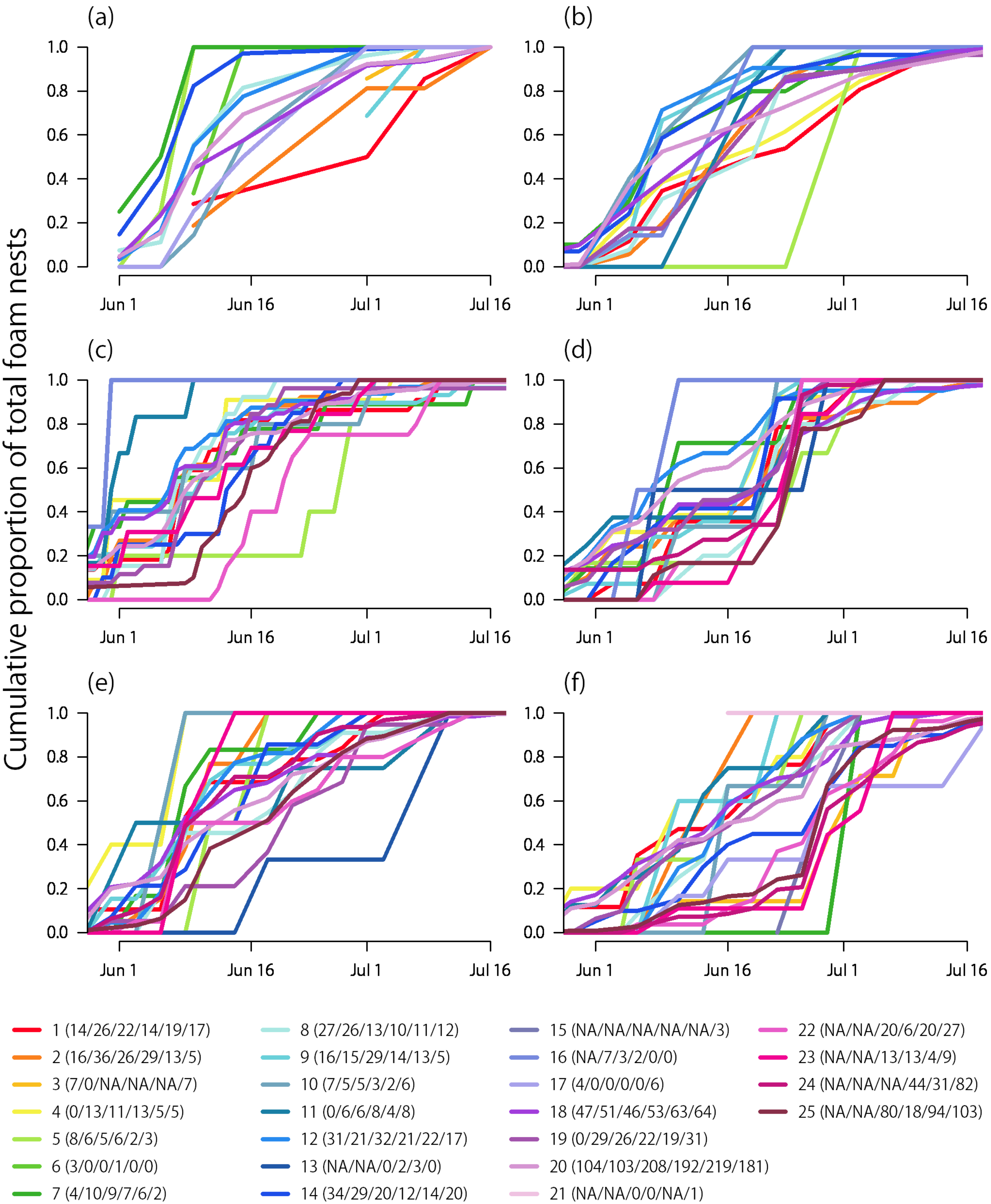
(a)
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Cumulative proportion of total foam nests
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0.2
0.0
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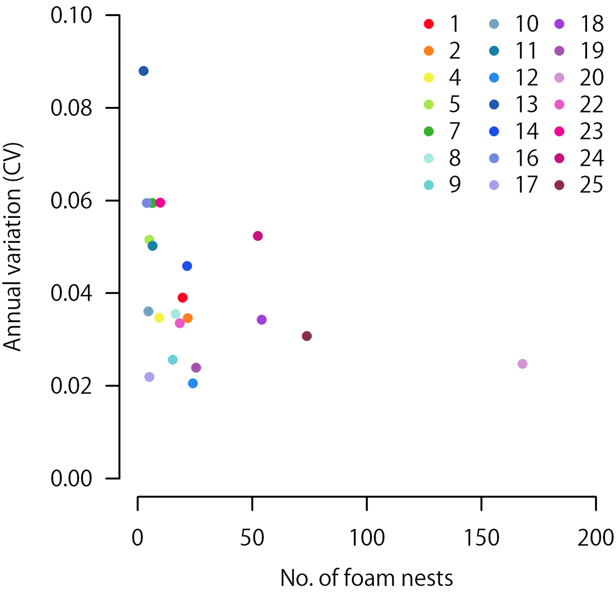
Jul 1
Jul 16
1 (14/26/22/14/19/17)
2 (16/36/26/29/13/5)
3 (7/0/NA/NA/NA/7)
4 (0/13/11/13/5/5)
5 (8/6/5/6/2/3)
6 (3/0/0/1/0/0)
7 (4/10/9/7/6/2)
8 (27/26/13/10/11/12)
9 (16/15/29/14/13/5)
10 (7/5/5/3/2/6)
11 (0/6/6/8/4/8)
12 (31/21/32/21/22/17)
13 (NA/NA/0/2/3/0)
14 (34/29/20/12/14/20)
15 (NA/NA/NA/NA/NA/3)
16 (NA/7/3/2/0/0)
17 (4/0/0/0/0/6)
18 (47/51/46/53/63/64)
19 (0/29/26/22/19/31)
20 (104/103/208/192/219/181)
21 (NA/NA/0/0/NA/1)
22 (NA/NA/20/6/20/27)
23 (NA/NA/13/13/4/9)
24 (NA/NA/NA/44/31/82)
25 (NA/NA/80/18/94/103)


Annual variation (CV)
0.10
0.08
0.06
0.04
0.02
0.00
0
50
100
150
200
No. of foam nests
1
2
4
5
7
8
9
10
11
12
13
14
16
17
18
19
20
22
23
24
25

Population Ecology

Spatial variation in breeding phenology at small spatial scales: a stochastic effect of population size

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Figure S1. *Rhacophorus arboreus* oviposits on the land.

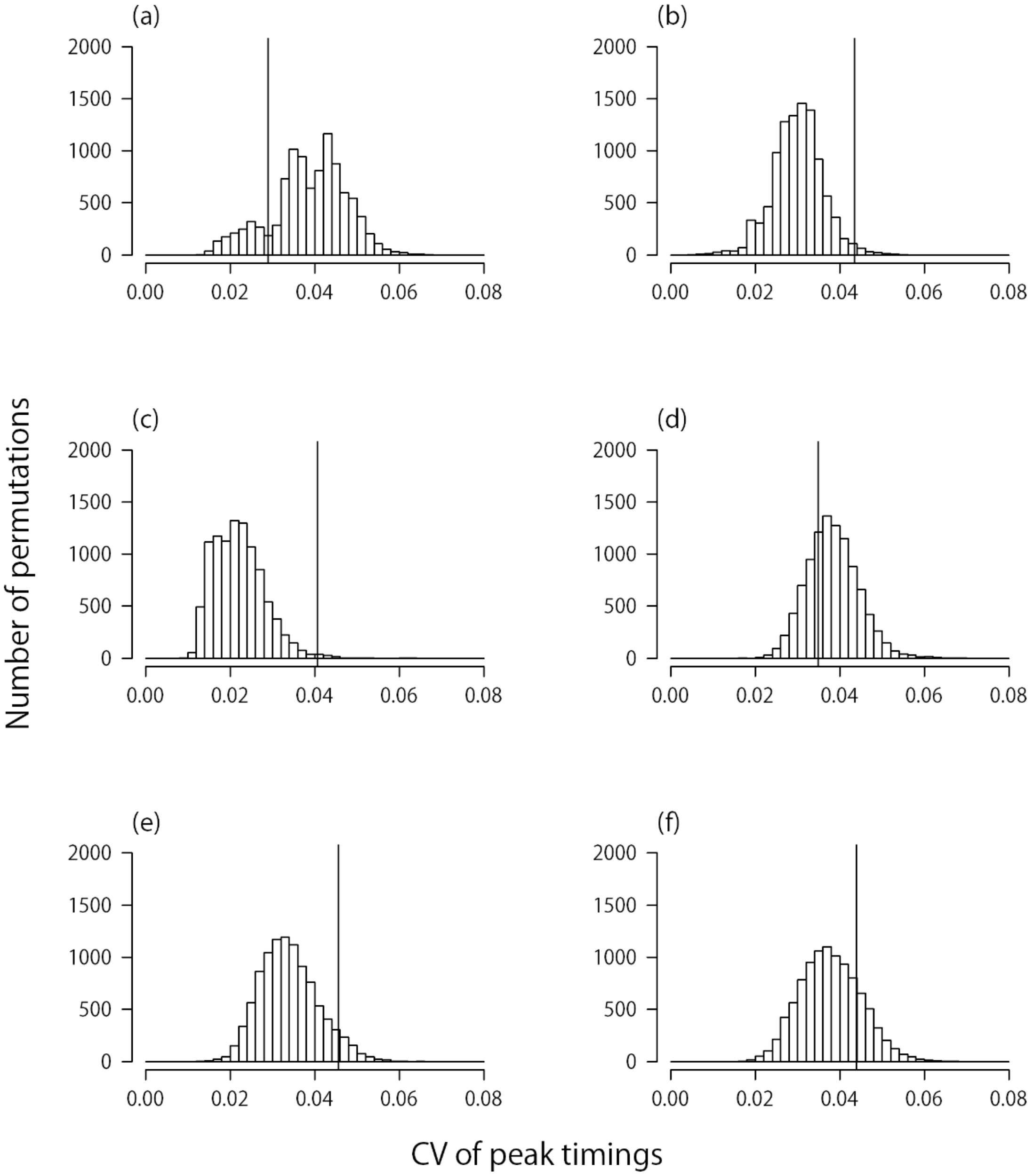

Figure S2. Distribution of the spatial variation in phenological peaks under the null hypothesis in (a) 2014, (b) 2015, (c) 2016, (d) 2017, (e) 2018, and (f) 2019. The x-axis represents the CV of the peak timings calculated across all populations. The y-axis represents the number of permutations. The black lines indicate the observed values of the CVs of peak breeding timing.

Table S1. Optimal accelerated failure time (AFT) model for the cumulative proportion of the total number of foam nests observed in each site in each year. In the AFT analysis, site 1 and the year 2014 were used as the contrasts against which the other sites and years were compared. Log (scale) denotes a scale parameter in the AFT model with a log-normal distribution.

		Estimate	Std. Error
Intercept		5.15	0.008
Year	2015	0	0.005
	2016	-0.04	0.005
	2017	-0.02	0.005
	2018	-0.03	0.005
	2019	0.01	0.005
Sites	2	0	0.009
	3	0.05	0.02
	4	-0.02	0.012
	5	0.01	0.015
	6	-0.01	0.036
	7	-0.04	0.013
	8	-0.01	0.01
	9	-0.01	0.01
	10	-0.02	0.015
	11	-0.03	0.014
	12	-0.03	0.009
	13	0.06	0.033
	14	-0.03	0.009
	15	0.04	0.042
	16	-0.04	0.022
	17	0.03	0.024
	18	-0.02	0.008
	19	0	0.009

	20	-0.02	0.007
	21	-0.03	0.071
	22	0.04	0.011
	23	0.02	0.013
	24	0.03	0.009
	25	0.03	0.008
Log (scale)		-2.64	0.013

Table S2. Results (*p*-values) of randomization tests in each site-year combinations.

Sites	2014	2015	2016	2017	2018	2019
1	-	0.013[a]	0.768	0.096	0.845	0.857
2	-	0.008[a]	0.926	0.012	0.483	0.847
3	-	-	-	-	-	-
4	-	0.077	0.792	0.381	0.697	0.873
5	0.699	0.006[a]	0.006[a]	0.017[a]	0.476	0.682
6	-	-	-	-	-	-
7	0.917	0.803	0.769	0.674	0.812	0.112
8	0.631	0.010[a]	0.809	0.133	0.177	0.865
9	-	0.749	0.458	0.411	0.473	0.876
10	0.217	0.655	0.606	0.223	0.821	0.794
11	-	0.240	0.977[b]	0.158	0.599	0.902
12	0.643	0.781	0.990[b]	0.966	0.812	0.855
13	-	-	-	0.371	0.000[a]	-
14	-	0.832	0.067	0.114	0.330	0.011[a]
15	-	-	-	-	-	-
16	-	0.141	0.91	0.816	0.816	-
17	0.150	-	-	-	-	0.134
18	0.017[a]	0.002[a]	0.972	0.001[a]	0.971	0.989[b]
19	-	0.015[a]	0.522	0.269	0.018[a]	0.854

20	0.000^{a}	0.977^{b}	0.881	0.999^{b}	0.004^{a}	0.998^{b}
21	-	-	-	-	-	-
22	-	-	0.001^{a}	0.021^{a}	0.295	0.002^{a}
23	-	-	0.100	0.001^{a}	0.691	0.005^{a}
24	-	-	-	0.000^{a}	0.464	0.000^{a}
25	-	-	-	0.000^{a}	0.003^{a}	0.000^{a}

Lowercase superscript letters ^{a} denotes that the median date of breeding was significantly later than that predicted by the null hypothesis ($\alpha < 0.025$), and ^{b} denotes that the median date of breeding was significantly earlier than that predicted by the null hypothesis ($\alpha > 0.975$). Hyphen denotes that the site-year combination was not used because of the small sample size.