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Nest-site selection by the harvest mouse *Micromys minutus* in seasonally changing environments

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We studied nest-site selection of harvest mice *Micromys minutus* (Pallas, 1771) for clarifying plant community characters suitable for nesting. We surveyed newly constructed nests in a marsh population and various plant characters likely to affect nest-site selection throughout the breeding season. The harvest mouce used common reed *Miscanthus sacchariflorus* and Japanese pampas grass *Miscanthus sinensis* more frequently than expected by the propotions of their areas occupied in summer, but shifted to *Carex confertiflora* after October. Although reed grass *Phragmites communis* was dominant in the marsh, mice did not choose this species. The 2 grass species selected in summer had a larger number of leaves per unit area, and the most selected species had a higher stem density. Both of these plant traits indicate a richness of nesting materials. The rapid increase in selection for *Carex* from autumn to winter was likely due to its leaf freshness and/or flexibility, as leaves of *Carex* maintained a high level of chlorophyll content until winter. The harvest mouse appeared to flexibly change its selection for nesting plants in response to seasonal environmental changes. We also discuss the implications of our findings in terms of harvest mouse habitat conservation.

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Introduction

The harvest mouse *Micromys minutus* (Pallas, 1771) is a small rodent that spends most of its life in tall grasslands, including marshes, pastures, and crop fields (Nordvig *et al*. 2001, Juškaitis and Remeisis 2007), where it constructs nests attached to stems of grasses (Harris 1979b, Dickman 1986, Churchfield *et al*. 1997, Giraudoux *et al*. 1998). Recently, such grasslands have been declining rapidly due to land use changes. In Japan, most of these habitats have been maintained by human activities such as cutting and burning or by temporal disturbances from flooding (Okubo 2002, Shoji 2003). Without such disturbances, grasslands tend to become forests because of plentiful rainfall. Therefore, it will be necessary to manage grasslands appropriately to preserve the habitats of the harvest mouse.

Although tall grasslands comprise many plant species, the harvest mouse uses a limited number of grass species for nesting (Shiraishi 1969, Morozumi and Morozumi 1996, Kuroda and Katsuno 2005, Surmacki *et al*. 2005). Plant architecture, such as density and height, is known to affect the nesting selections of this mouse (Surmacki *et al*. 2005). However, other factors may potentially be concerned, as nests are occasionally constructed in low vegetation, such as fox tail *Setaria viridis* and *Carex* (Sawabe *et al*. 2005). Furthermore, soil water content, an index of flooding, may also affect mouse selectivity (Morozumi and Morozumi 1996, Surmacki *et al*. 2005). To manage plant communities effectively that are favorable for the harvest mouse, it is necessary to explore which factors affect nesting selections under various conditions.

Here, we examined the seasonal changes in and the factors affecting harvest mouse nest-site selections in a marsh surrounded by secondary deciduous forest, which is a common habitat for the harvest mouse in Japan. The study site was characterized by a gradient from permanent wet to dry microhabitats and contained a variety of plant species that differ in their phenology. These characteristics may aid in understanding the mechanisms that influence context-dependent nest-site selections of the harvest mouse. We predicted that density and height of plants are the primary determinants of nest-site selection but the degree of selection is changed by seasonal die-back of plants as well as wetness of the soil.

Material and methods

The study was conducted in a marsh located at Sayama Hill, central Japan. The hill is approximately 35 km^2, contains a reservoir at its center, and is primarily deciduous forest. This area also has several small valleys with patches of grassland. The study marsh is approximately 3 ha and is dominated by reed grass *Phragmites communis*, common reed *Miscanthus sacchariflorus*, *Carex confertiflora*, common cattail *Typha latifolia*, and water pepper *Polygonum thunbergii*. In addition, communities of Japanese pampas grass *Miscanthus sinensis* surround the marsh.

Harvest mouse nests were observed once a week throughout the breeding season from June to December in 2003 and 2004. We established five 200-m census routes in the marsh and counted newly emerging nests. This census appeared to cover all nests in the marsh. We marked new nests on the stem near the nests and measured the heights of nests and the plants where nests were found. The number of new nests found within each plant community was summed monthly. The area of each plant community was measured and summed in all across the entire marsh. We examined the architecture and phenology of four plant species (reed grass, common reed, Japanese pampas grass, and *Carex*) 3 times a year (except for plant height and stem density) in July, October, and December. To measure plant height, we established five 50 × 50-cm quadrats for each species and measured foliage height monthly. To estimate the vertical distribution of leaf densities, we set out five 33 × 33-cm quadrats for each species, removed all leaves, and measured the sum of leaf areas within every 50-cm vertical section aboveground in the laboratory. We measured soil water content at five randomly selected locations within each plant community, using a water content sensor (Campbell Scientific, Inc). We also measured chlorophyll contents of 10 leaves of each species to obtain the index of freshness and greenness of leaves, using a chlorophyll sensor (Minolta Spad-502).

For estimations of mouse nest-site selectivity, we conducted a binomial test (sequential Bonferroni-corrected) to detect differences between the actual and expected number of nests found in each plant community, based on the proportion of the area occupied by that plant community within the marsh. To test for differences in leaf area, plant height, chlorophyll content, and soil water content, we conducted a two-way analysis of variance (ANOVA) using plant species and month as fixed factors. When interactions were significant, we performed one-way ANOVAs for each month, using plant species as factors, followed by Tukey's multiple comparisons. For leaf area, we used the largest value among the vertical layers for each species.

Results

During the breeding season, we found a total of 92 nests in 2003 and 148 in 2004. There was a strong correlation between nest heights and the plant heights where nests were found ($R^2 = 0.954$, $p < 0.05$, $n = 119$). The nest heights were approximately four-fifth of the plant height. Figure 1 presents changes in nest-site selections for a particular plant species expressed as the ratio between the number of nests found on each plant species and the number expected based on the area occupied by that species. The harvest mouse selectively used common reed *Miscanthus sacchariflorus* and Japanese pampas grass *Miscanthus sinensis* for nest sites until Septem-

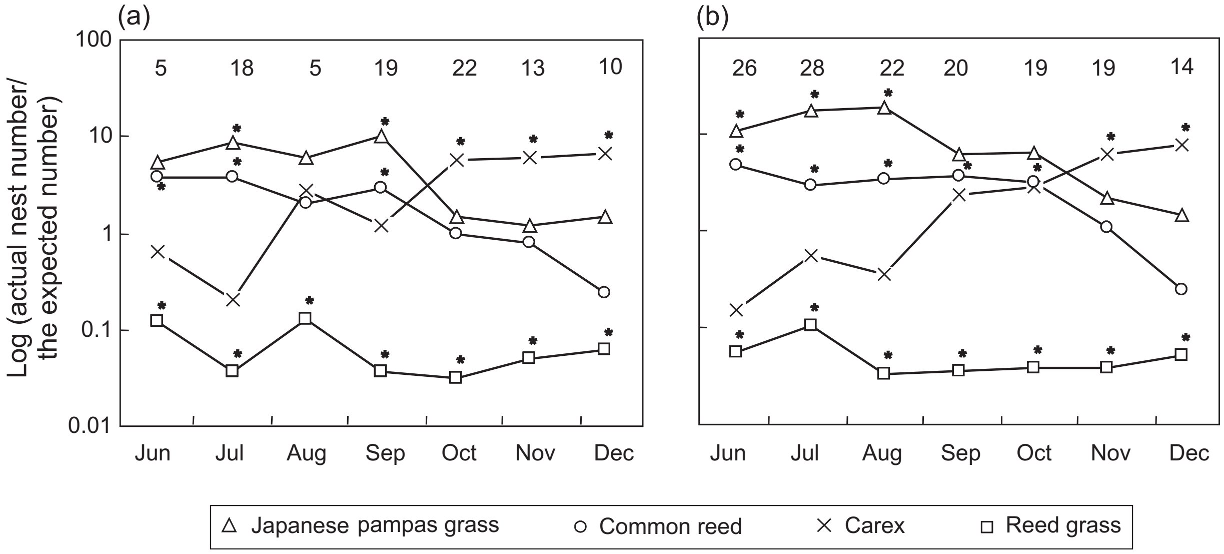

Fig. 1. Nest-site selection for plant species by harvest mice *Micromys minutus* in 2003 (a) and 2004 (b). Numerals above the panels were the total number of nests of 4 communities newly found in each month. Asterisks denote significant differences from the expected values (1.0).

ber or October, after which it used *Carex* most frequently (Fig. 1). Reed grass was not selected throughout the breeding season (Fig. 1). These patterns were consistent in both years.

The two-way ANOVA revealed significant differences in leaf area between the 4 species ($F_{3,6} = 96.99$, $p < 0.01$), between months ($F_{2,6} = 5.21$, $p = 0.049$), and their interaction ($F_{3,48} = 3.80$, $p < 0.01$). The subsequent one-way ANOVAs indicated a significant difference in leaf area among the four species in July ($p < 0.05$), with reed grass exhibiting the lowest leaf area and *Carex* exhibiting the highest (Fig. 2a). The 3 grasses (reed grass, common reed, and Japanese pampas grass) lost their leaves in autumn, thus leaf area in these species declined markedly at this time (Fig. 2a).

Soil water content differed significantly among communities of the 4 species (two-way ANOVA, $F_{3,6} = 10.41$, $p < 0.01$), with the lowest values in Japanese pampas grass and common reed. Soil water content in reed grass and *Carex* communities remained greater than 100% throughout the season (Fig. 2b).

A two-way ANOVA revealed no significant differences in chlorophyll content among the 4 species ($F_{3,6} = 3.76$, $p = 0.08$), but chlorophyll clearly differed between months ($F_{2,6} = 35.36$, $p < 0.01$). The amount of chlorophyll in the 3 grasses declined after October, while *Carex* maintained nearly the same level throughout the year (Fig. 2c). This pattern can be explained by the fact that *Carex* remained green and fresh until December, while the grass species blossomed in September and withered after November. Japanese Pampas grass showed the highest stem density and *Carex* showed the lowest (Fig. 2d), with no obvious seasonal variation. Community heights changed seasonally except for *Carex* (Fig. 2e).

Discussion

At our study site, the harvest mouse did not select reed beds for nest-sites throughout the breeding season, despite the fact that reed grass was the dominant species. This result was consistent with previous findings (Miyahara 1995, Morozumi and Morozumi 1996). In contrast, common reed was used frequently, even though it grows sympatrically with reed grass. These two species exhibit similar heights and growth processes, but common reed had a much larger

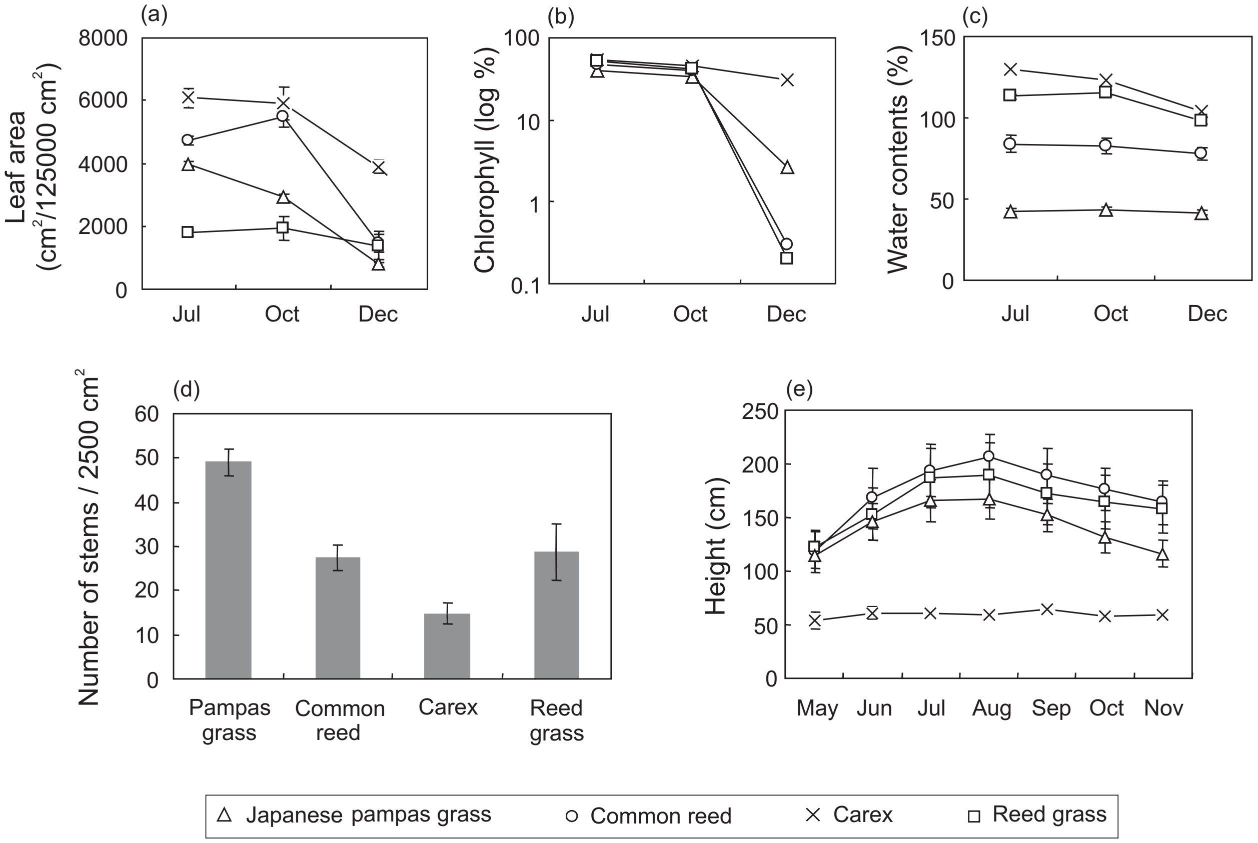

Fig. 2. Characters and microhabitat of plant species available for nest-site of harvest mice *Micromys minutus* (mean ± SE). (a) Leaf area, (b) Chlorophyll, (c) Soil water content, (d) Stem density and (e) Community height.

leaf area per unit area. Bence *et al*. (2003) surveyed the community structure of various plant species and reported that harvest mice selected dense vegetation for nesting, probably because a certain number of leaves is necessary for nest construction. The low selection for reed grass is likely due to its smaller number of leaves per area, making it difficult for the harvest mouse to construct nests.

Our results also indicated that nest-site selection changed seasonally. While the harvest mouse did not select reed grass at any time of year, its selection for the other three species changed in October. The harvest mouse used *Carex* more frequently than common reed and Japanese pampas grass in November and December. Plant height, an important factor for nest construction (Sawabe *et al*. 2005), was unlikely related to this shift, as Japanese pampas grass and common reed were still taller than *Carex* in October and later months. This seasonal shift was probably due to differences in the phenologies of these plant species. Because the harvest mouse tears and weaves leaves for nesting, the leaves must be flexible for nest construction. Therefore, *Carex* was likely selected because only its leaves remained green and fresh even in December, suggesting that *Carex* may be an alternative host plant for the harvest mouse when other species are not suitable, although the community height was low.

The harvest mouse exhibited the strongest selection for Japanese pampas grass at our study site. The number of leaves and height of Japanese pampas grass was intermediate, and its seasonal change was similar to that of common reed. However, the stem density of Japanese pampas grass was the highest among the four species, potentially facilitating nest construction. Differences in microhabitats may be another reason why Japanese pampas grass was selected. This species grows in dry environments with no flooding throughout the year, while common reed and *Carex* are found primarily in wet microhabitats. According to Shiraishi (1969), the harvest mouse builds nests in rice fields after water is extracted. Also, the harvest mouse were never observed within 8 months after a flood (Wijnhoven *et al*. 2005). Therefore, the harvest mouse's selection for Japanese pampas grass was likely due in part to the drier microhabitat characteristics.

Our results indicated that nest-site selection by the harvest mouse was influenced not only by the height and density of vegetation (Bence *et al*. 2003), but also by other plant architectural features as well as microhabitat conditions. In addition, we have demonstrated that the harvest mouse flexibly shifts among plant species for nest sites depending on the season. Our findings indicate that for habitat conservation of the harvest mouse, it is not sufficient to merely conserve grasslands. Attention must be paid to the specific plant species and their characteristics when applying conservation measures. For example, in Japan, high priority should be given to common reed and Japanese pampas grass conservation relative to reed grass. Moreover, the conservation of a *Carex* community may extend the harvest mouse breeding season into winter. Although the length of the breeding season is often thought to be related to temperature and precipitation (Shiraishi 1962, Harris 1979a), the effects of the phenology of local vegetation have yet to be considered. The influence of plant phenology on the harvest mouse breeding season could be examined by comparing the length of the breeding season in local populations differing in plant species composition. Furthermore, to make appropriate habitat management decisions for the harvest mouse, it is necessary to determine the extent to which the length of the breeding season affects harvest mouse population dynamics.

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