

Teruyoshi Nagamitsu · Tanaka Kenta
Naoki Inari · Etsushi Kato · Tsutomu Hiura

Abundance, body size, and morphology of bumblebees in an area where an exotic species, *Bombus terrestris*, has colonized in Japan

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Abstract An exotic bumblebee species, *Bombus terrestris*, has colonized in Japan and becomes dominant in some local communities. We examined the effects of land use and bumblebee abundance on the number and body size of bumblebees collected using window traps in a lowland area in the southern Ishikari district, Hokkaido. In 2004, we collected 922 bumblebees of six species using 70 traps at 17 sites. A statistical model fitted to the data demonstrated that dispersion from commercial *B. terrestris* colonies used in greenhouses positively affected the number of *B. terrestris* caught by each trap. This exotic species was abundant in sites where paddy fields were prevalent, but three native species, *B. hypocrita*, *B. ardens*, and *B. diversus*, were abundant in sites where farms and woodlands were widespread. The local abundance of *B. terrestris* was not associated negatively with the number and body size of native bumblebees. Thus, we

did not find any competitive interactions between exotic and native bumblebees although habitat conditions seem to be common determinants of the bumblebee populations. A morphological analysis showed that *B. terrestris* had intermediate tongue length between *B. hypocrita* and *B. ardens*.

Keywords Biological invasion · *Bombus terrestris* · Competition · Landscape · Window trap

Introduction

Bombus terrestris (L.) (Apidae) had become established in the wild in Japan until 1996, following escapes from commercial colonies used for pollination of tomato crops in greenhouses since 1992 (Matsumura et al. 2004). Invasions of *B. terrestris* have occurred mainly in cultivated areas (Nakajima et al. 2004). In some lowland agricultural areas in Hokkaido, *B. terrestris* has become dominant in local bumblebee communities (Inari et al. 2005). According to the spatial patterns of invasions, it is not clear whether the dominance of *B. terrestris* has resulted from competitive exclusion of native species by *B. terrestris* or not, because it is possible that *B. terrestris* has colonized without reducing populations of the native species in the agricultural areas where the native species were originally rare. However, the abundant *B. terrestris* potentially competes with the native species by exploiting common resources.

Competition for food and nest sites is to be expected because exotic social bees are likely to consume a substantial fraction of the available resources of native bees (Goulson 2003b). In Tasmania, the competitive effects of *B. terrestris* on native bees, *Chalicodoma* spp. (Megachilidae), were demonstrated when the native bees visited flowers of *Gompholobium huegeli* Benth. (Fabaceae) (Hingston and McQuillan 1999). Such competition may decrease the populations of native species. On the basis of records of sighting of pollen-collecting bumblebees, breeding populations of *B. terrestris* were established

T. Nagamitsu (✉)
Hokkaido Research Center,
Forestry and Forest Products Research Institute (FFPRI),
Sapporo 062-8516, Japan
E-mail: nagamit@ffpri.affrc.go.jp
Tel.: +81-11-8514131
Fax: +81-11-8514167

T. Kenta
Department of Animal and Plant Sciences,
University of Sheffield,
Sheffield S10 2TN, UK

N. Inari
Institute of Low-Temperature Science,
Hokkaido University,
Sapporo 060-0819, Japan

E. Kato
Graduate School of Environmental Earth Science,
Hokkaido University,
Sapporo 060-0810, Japan

T. Hiura
Tomakomai Research Station,
Field Science Center for Northern Biosphere,
Hokkaido University,
Tomakomai 053-0035, Japan

throughout most of Tasmania (Hingston et al. 2002). A survey using quantitative searches showed that *B. terrestris* was far more abundant in gardens and arable fields than in native vegetation (Goulson et al. 2002). Furthermore, the abundance and diversity of native flower-visiting insects did not differ between sites in which *B. terrestris* was absent and present although their diversity differed among habitat types (Goulson et al. 2002). Thus, not only competitive interactions but also habitat conditions potentially affect bumblebee abundance.

Food supply is a major determinant of the body size of bumblebee workers (Sutcliffe and Plowright 1988). Thus, a reduction in body size indicates shortage of food. In a field experiment in alpine meadows, the removal of potential competitors increased the body size accompanied by a shift in flower use, which supports exploitative competition for food (Bowers 1985b). On the other hand, the body size of workers has been known to change seasonally and vary spatially (Inoue and Kato 1992; Tomono and Sota 1997). If the body size of native bumblebees decreases as the abundance of *B. terrestris* increases, interspecific competition for food is suggested as long as the effects of other factors on body size are discriminated from the competitive effects.

In this study, we collected bumblebees using window traps in various landscapes in an area where *B. terrestris* has colonized. We examined the effects of habitat conditions characterized by land use and the local abundance of exotic and native bumblebees on the number and body size of trapped bumblebees using statistical models. The morphology of exotic and native bumblebees was also analyzed because morphological characters are closely related to floral resource use.

Materials and methods

Study area

This study was conducted in the southern Ishikari district, Hokkaido, Japan (42°50–58'N and 141°32–44'E; Fig. 1). The study area was the lowland at 6–93 m

altitudes. In the study area, land use consisted of farms, including crop fields and pastures (36%), woodlands (22%), urban areas, including residential and industrial areas (14%), paddy fields (14%), and others such as abandoned fields, barren lands, bare lands, roads, and water surface (14%); this information was based on the latest topographic maps in the 1:25,000 scale published from Geographical Survey Institute of Japan. According to Inari et al. (2005), 350 commercial colonies of *B. terrestris* were used in 33 greenhouses in the study area during 2003 (Fig. 1).

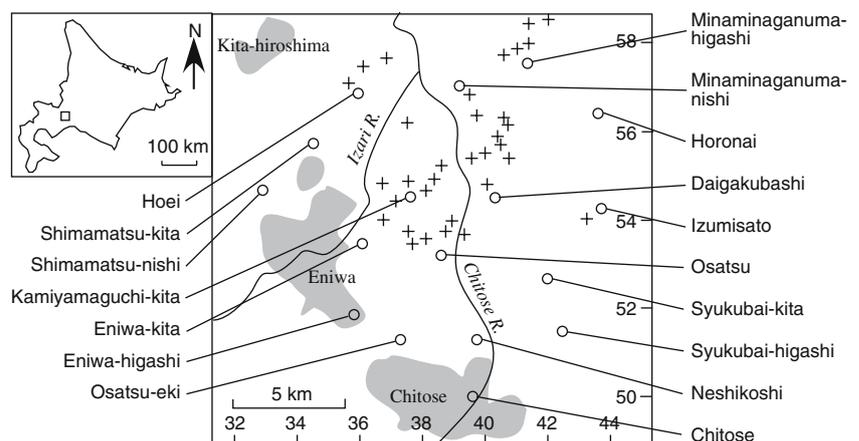
Trapping

Several belts of windbreak forests run through the study area. In 2004, 17 trapping sites were located in the windbreak forests surrounded by various landscapes (Fig. 1, Appendix 1). At each site, we set two or three traps, both at the canopy and the understory. Each canopy trap was suspended using a rope through a pulley fixed to a canopy branch. Each understory trap was tied directly to a tree branch. The height of the canopy traps ranged from 6.8 to 13.3 m depending on the canopy height of the forests, and that of the understory ones ranged from 0.9 to 1.7 m (Appendix 1). The trap used was a kind of interception trap with a roof, perpendicularly crossed transparent vanes, and a plastic white tray (Nakashizuka 2002).

Two and four liters of water with a small amount of odorless detergent were poured into the trays of the understory and canopy traps, respectively. Bumblebees that collided with the transparent vanes fell into the water. One week after the addition of water, the trapped bumblebees were collected from the tray, and the water was then discarded. Trapping was conducted nine times at intervals of about 2 weeks between 2 June and 22 September in 2004.

In order to examine the variation in the number of bumblebees of each species collected using the traps, we set understory traps at a site (100 × 20 m area in a windbreak forest) for a week in three periods: 18 traps at Neshikoshi between 26 July and 1 August 2003; 21 traps

Fig. 1 Trapping sites (circles) and greenhouses (crosses) in which commercial *Bombus terrestris* colonies were used in the study area in Hokkaido. Urban areas of three cities (shaded areas) and two rivers (lines) are also shown. The horizontal and vertical axes show the longitude (141°32–58'N) and latitude (42°50–58'N), respectively



at Kamiyamaguchi-kita between 16 and 22 August 2003; and 22 traps at Kamiyamaguchi-kita between 16 and 22 July 2004 (Appendix 1). The frequency distributions of the number of bumblebees of each species collected from a trap during a week were not significantly different from the poisson distributions (Kolmogorov–Smirnov tests for the three trapping examinations: $P > 0.816$, $P > 0.975$, and $P > 0.811$). Thus, the number of trapped bumblebees was regarded to follow a poisson distribution.

Morphology

Trapped bumblebees were preserved in 99.5% ethanol and classified into species and sexes. In the following analysis, the four abundant species, *B. terrestris*, *B. hypocrita*, *B. ardens*, and *B. diversus* were examined. As an indicator of body size, the head width (HW) of trapped bumblebees was measured using an electric digital caliper (CD-20, Mitsutoyo Coporation, Kawasaki). On the basis of the gap in the HW of females trapped on each trapping date, the females were classified into queens and workers.

The morphology of 12 queens and 12 workers randomly sampled from the trapped females of each of the four abundant species was measured under a binocular microscope with a micrometer (SMZ-U, Nikon, Tokyo). Seven characters related with foraging behavior were selected according to Inoue and Kato (1992): HW, head length (HL), distance between the bases of mandibles (MW), prementum length (PL), glossa length (GL),

marginal cell length (ML) of the fore wing, and corbicular length (CL) of the hind leg (Fig. 2).

The HW measured using a digital caliper (HW') was about 5% smaller than that measured under a microscope ($HW' = HW \times 0.949 \pm 0.003$ (estimated coefficient \pm SE), $r^2 = 0.999$, $n = 96$).

In order to examine the variation in the HW among females, we measured the HW' of 110 queens of *B. terrestris* collected in Neshikoshi on 3 May 2005. The frequency distribution of the HW' of *B. terrestris* queens was not significantly different from a normal distribution (Kolmogorov–Smirnov test, $P > 0.610$). Thus, the HW' was regarded to follow a normal distribution.

Data analysis

The foraging ranges of *B. terrestris* colonies were less than 625 m (Darvill et al. 2004) and less than 758 m (Knight et al. 2005) in farmlands. Thus, bumblebees collected in a trap could come from colonies within about a 0.75-km radius around the trap, and the foraging areas of the colonies covered a 1.5-km radius around the trap. Because the 17 trapping sites were apart from each other at more than 2.7 km distances, samples collected from the 17 sites seem to be regarded as statistically independent samples.

In order to summarize land use, a principal component analysis was performed for the 17 sites using four variables: the percentages of paddy field area, farm area, urban area, and woodland area within a 1.5-km radius around each site (Appendix 1). The first, second, and third principal components explained 59.1, 26.6, and 13.5%, respectively, of the total variance of the four land use variables. The first principal component, p_1 , reflected urbanization because the urban area contributed positively but the farm and paddy field areas contributed negatively to p_1 (Fig. 3). The second principal component, p_2 , indicated prevalence of paddy fields in farmlands because the paddy field area contributed positively but the farm and woodland areas contributed negatively to p_2 (Fig. 3).

The total number of bumblebees of each species, N_i , collected during 2004 from the i th trap was assumed to follow a poisson distribution with the mean, λ_i :

$$N_i \sim \text{Poisson}(\lambda_i).$$

We let a linear predictor determine a logarithmic link function of the mean, λ_i ,

$$\log_e \lambda_i = B + L + H + A + L : A + H : A$$

where B is the background effect, L is the land use effect, H is the effect of the trap height, and A is the effect of the local abundance of other species. $L:A$ and $H:A$ are interactions between L and A as well as between H and A , respectively.

In the three native species, the background effect was a constant (intercept),

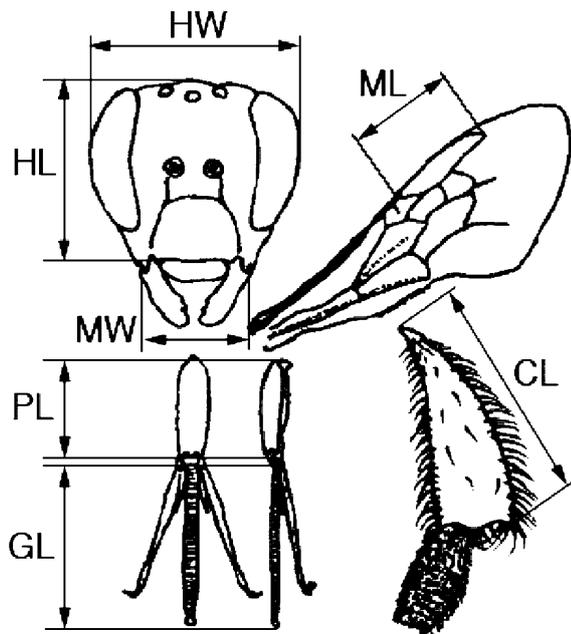


Fig. 2 Morphological measurements for bumblebees; HW head width, HL head length, MW distance between the bases of mandibles, PL prementum length, GL glossa length, ML marginal cell length of the fore wing, and CL corbicular length of the hind leg

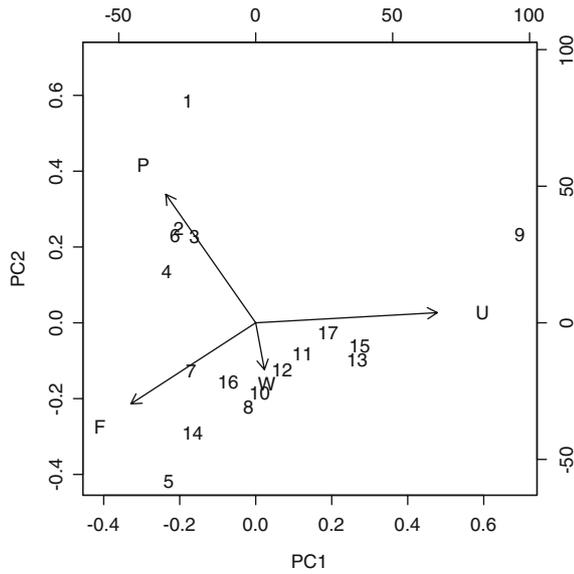


Fig. 3 Principal component analysis on 17 trapping sites (1–17; Appendix 1) using four variables: the percentages of paddy field area (P), farm area (F), urban area (U), and woodland area (W) within a 1.5-km radius around the sites. *Arrows* show the vectors of the loading factors for the first and second principal components ($PC1$ and $PC2$) in the four variables

$$B = \beta_0.$$

In *B. terrestris*, dispersion from commercial colonies in 33 greenhouses was used to determine the background effect in the formula:

$$B = \beta_0 \sum_{j=1}^m s_j f(\delta, d_{ij}),$$

where β_0 is a constant, s_j is the number of commercial colonies used in the j th greenhouse ($m = 33$), and f is a diffusion function (two-dimensional normal distribution):

$$f(\delta, d_{ij}) = \frac{1}{2\pi\delta^2} \exp\left(-\frac{d_{ij}^2}{2\delta^2}\right),$$

where δ is a diffusion coefficient (the standard deviation of the two-dimensional normal distribution) and d_{ij} is the distance (km) between the i th trap and the j th greenhouse.

The land use effect consisted of $p1_i$ and $p2_i$ of the tapping site of the i th trap, which were obtained from the principal components of land use (Fig. 3):

$$L = \beta_{p1}p1_i + \beta_{p2}p2_i.$$

The height (m), h_i , of the i th trap (Appendix 1) was used to determine the effect of trap height,

$$H = \beta_h h_i.$$

The mean number of bumblebees collected from traps at the same site of the i th trap (Appendix 2) was re-

garded as the local abundance. For *B. terrestris*, the local abundances of the three native species were summed up (n_i), which was used to determine the effect of the local abundance of other species,

$$A = \beta_n n_i.$$

For the three native species, the local abundance of *B. terrestris*, e_i , and the total abundance of other two native species, n_i , were used to determine the effect,

$$A = \beta_e e_i + \beta_n n_i.$$

Finally, the interactions for *B. terrestris* were

$$L : A + H : A = \beta_{p1:n}p1 : n_i + \beta_{p2:n}p2 : n_i + \beta_{h:n}h : n_i,$$

and those for the three native species were

$$L : A + H : A = \beta_{p1:e}p1 : e_i + \beta_{p2:e}p2 : e_i + \beta_{h:e}h : e_i + \beta_{p1:n}p1 : n_i + \beta_{p2:n}p2 : n_i + \beta_{h:n}h : n_i.$$

The HW' (mm) of workers of each species, W_{it} , collected from the i th trap on the t th date was assumed to follow a normal distribution with the mean, μ_{it} , and the standard deviation, σ :

$$W_{it} \sim \text{Normal}(\mu_{it}, \sigma^2).$$

We let a linear predictor determine the mean, μ_{it} ,

$$\mu_{it} = \beta_0 + S + L + H + A' + L : A' + H : A',$$

where β_0 is a constant (intercept), S is the seasonal effect, L is the land use effect, H is the effect of the trap height, and A' is the effect of the local abundance of the four species. $L:A'$ and $H:A'$ are interactions. The effects of L and H are the same as above.

The date (the number of days after 1 January 2004), t , when workers were collected was used to determine the seasonal effect,

$$S = \beta_t t.$$

The local abundance, the same as above, was calculated for *B. terrestris* (e_i) and the total of three native species (n_i) to determine the effect of the local abundance,

$$A' = \beta_e e_i + \beta_n n_i.$$

Finally, the interactions were

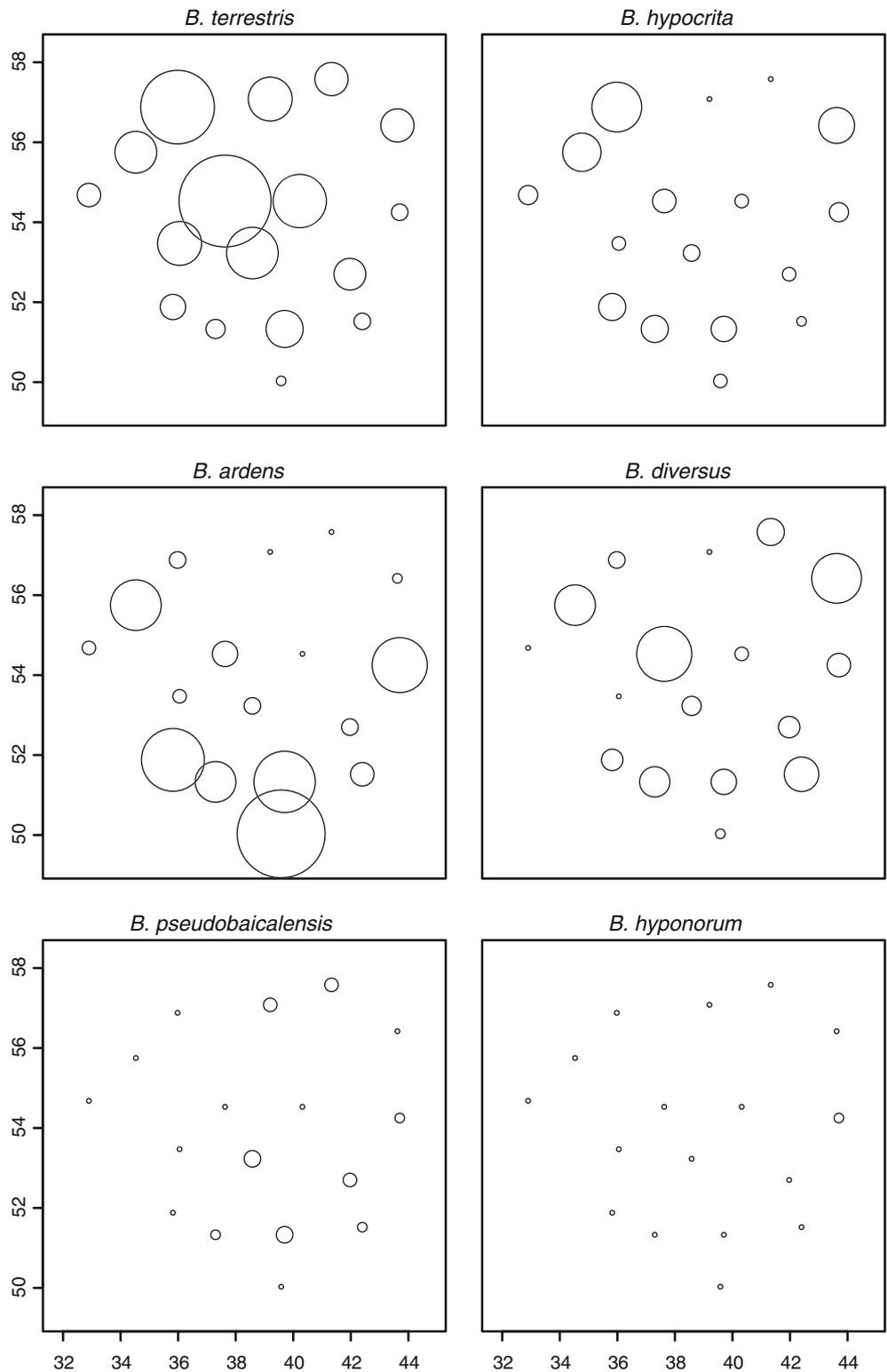
$$L : A' + H : A' = \beta_{p1:e}p1 : e_i + \beta_{p2:e}p2 : e_i + \beta_{h:e}h : e_i + \beta_{p1:n}p1 : n_i + \beta_{p2:n}p2 : n_i + \beta_{h:n}h : n_i.$$

The independent variables, $p1$, $p2$, h , e , n , t , except for the intercepts were standardized by subtracting the mean from each variable and dividing by the standard deviation to reduce collinearity between the variables and their interactions (Quinn and Keough 2002). The models were evaluated based on Bayesian information criterion (BIC) (Burnham and Anderson 2002) using *R* 2.1.0 statistical software (*R* Development Core Team 2003).

The model with the lowest BIC was selected in a process of stepwise selection. In the stepwise process, the parameters were removed from the models as long as the BIC decreased. Maximum likelihood estimates of the parameters, β s, were obtained in the selected models.

Null hypotheses that parameters estimated in the selected models were equal to 0 were tested using Wald's t statistics. In order to estimate the diffusion coefficient, δ (km), we calculated BIC of the abundance model of *B. terrestris* for various δ values ranging from 1.0 to 16.0 at

Fig. 4 Number of bumblebees (indicated by the area of circles) of six species trapped at 17 sites. The horizontal and vertical axes show the longitude (141°32–44'N) and latitude (42°50–58'N), respectively



0.1 intervals. The δ value that yielded the lowest BIC was selected.

A principal component analysis for 96 females sampled from the four abundant species was performed on the basis of the seven morphological characters. A canonical discriminant analysis to discriminate among the four species was also conducted. Some characters that represented morphological variation were selected according to the two multivariate analyses, and the relationships among the characters were compared among the four species.

Results

Abundance

In 2004, 922 bumblebees of six species were collected from 70 traps at the 17 sites, and each trap caught an average of 13.01 bumblebees (Appendix 2). Among the six species, *B. terrestris* was the most abundant (353 individuals; 128 queens, 187 workers, and 38 males), followed by *B. ardens sakagami* Tkalcu (295; 53, 192, and 50), *B. diversus tersatus* Smith (147; 61, 80, and 6), *B. hypocrita sapporoensis* Cockerell (110; 19, 82, and 9), *B. pseudobaicalensis* Vogt (16; 10, 4, and 2), and *B. hyponorum koropokkrus* Sakagami et Ishikawa (1; 0, 1, and 0).

Spatial distributions of the number of bumblebees trapped at the 17 sites showed different patterns among the four abundant species (Fig. 4). *B. terrestris* was frequently trapped at the center of the agricultural area, whereas *B. ardens* was abundant at the periphery of the lowland. *B. hypocrita* and *B. diversus* were scattered throughout the study area, and their abundance varied among the sites.

The dispersion of *B. terrestris* from commercial colonies in 33 greenhouses was fitted to statistical models.

Based on a model selected, the estimate of the diffusion coefficient, δ , was 7.3 km, and the dispersal curve was obtained.

In a model fitted to the number of trapped *B. terrestris*, parameters of the dispersion, paddy field prevalence, and trap height were significantly positive, and a parameter of the urbanization was significantly negative (Table 1). The local abundance of native species had significant interactions with the land use and trap height. Parameters of these interactions indicated that an increase in the local abundance of native species intensified the effects of land use and trap height in *B. terrestris*.

Models with significantly negative parameters of the paddy field prevalence were fitted to the number of trapped bumblebees of all the three native species (Table 1). In addition, a significantly positive parameter of the urbanization was estimated in *B. ardens*. The trap height had significantly positive parameters in *B. hypocrita* and *B. ardens*. In *B. ardens*, there was a synergistic effect of the trap height and the local abundance of other native species. The local abundance of *B. terrestris* had significantly positive parameters in *B. hypocrita* and *B. diversus*.

Body size

On most trapping dates, queens and workers were clearly discriminated based on their HW' (Fig. 5). Trapped queens included old ones that emerged in 2003 and new ones that emerged in 2004. Old queens hibernated and then became active in the period of colony foundation, and new queens appeared at the end of the colony growth period. The periods when old and new queens were trapped were continuous in *B. terrestris* and *B. ardens* but intermittent in *B. hypocrita* and *B. diversus* (Fig. 5). The period of worker appearance was longer in

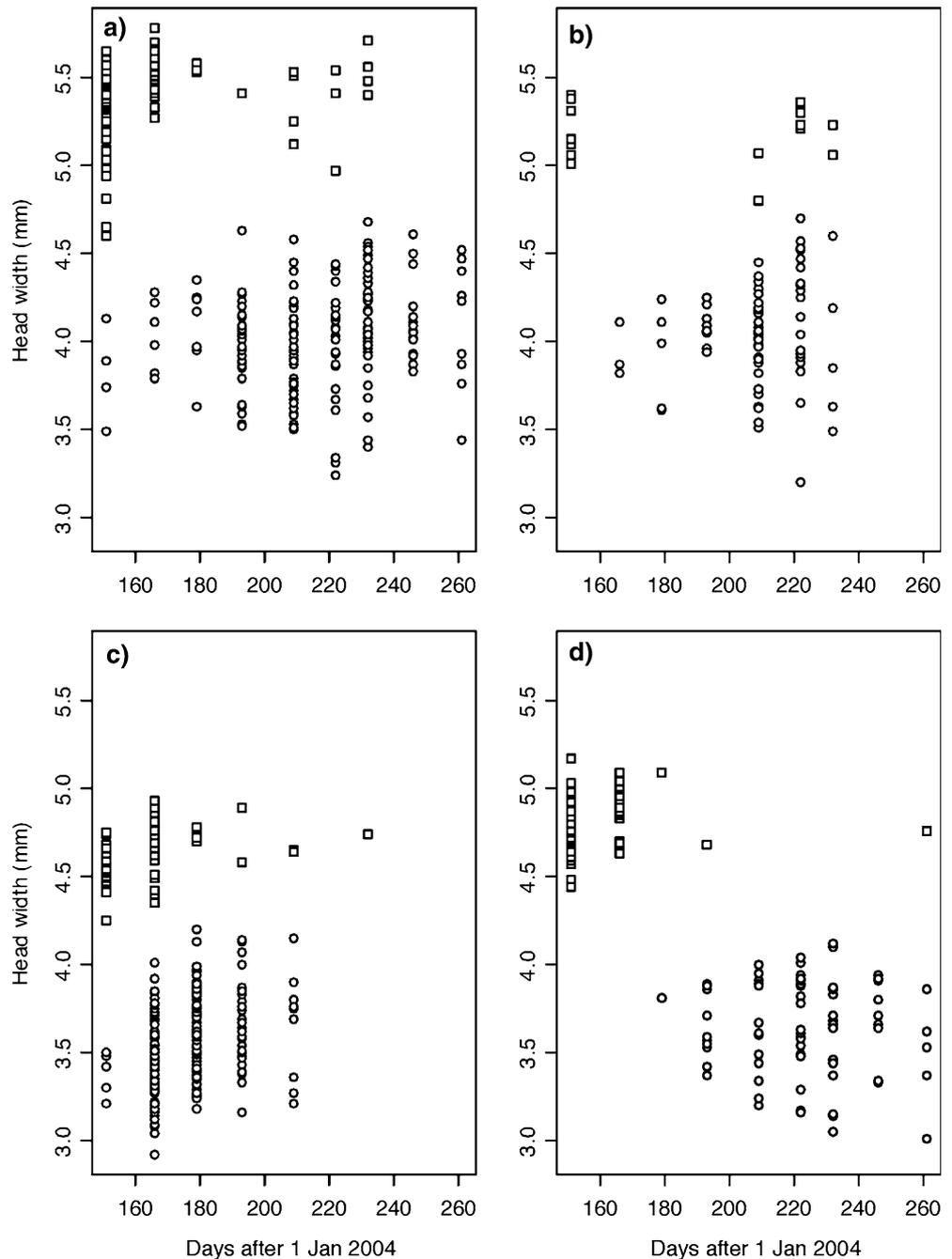
Table 1 Parameter estimates in generalized linear models for the number of trapped bumblebees of four abundant species

Parameter	Estimate in selected model for the number of trapped bumblebees of			
	<i>Bombus terrestris</i>	<i>B. hypocrita</i>	<i>B. ardens</i>	<i>B. diversus</i>
Dispersion or intercept	1.706***	0.171	0.682***	0.486***
Land use				
Urbanization (<i>p</i> 1)	-0.656***	NS	0.964***	NS
Paddy field prevalence (<i>p</i> 2)	0.458***	-0.340**	-0.635***	-0.752***
Trap height (<i>h</i>)	0.586***	0.656***	0.673***	NS
Local abundance				
<i>B. terrestris</i> (<i>e</i>)	-	0.426***	NS	0.672***
Other native species (<i>n</i>)	0.023	NS	0.147	NS
Interactions				
<i>p</i> 1: <i>n</i>	-0.342***	NS	NS	NS
<i>p</i> 2: <i>n</i>	0.321***	NS	NS	NS
<i>h</i> : <i>n</i>	0.228***	NS	0.200*	NS

-: parameters are not included in the generalized linear model, NS parameters are not selected in the generalized linear model with the lowest BIC (other interactions were not selected)

*0.01 ≤ *P* < 0.05; **0.001 ≤ *P* < 0.01; ****P* < 0.001 (Wald's *t* statistics: parameters different from zero)

Fig. 5 Seasonal changes in the HW' (mm) of females of **a** *B. terrestris*, **b** *B. hypocrita*, **c** *B. ardens*, and **d** *B. diversus*. Squares and circles indicate queens and workers, respectively



B. terrestris than in the three native species. Among the native species, *B. ardens* workers appeared the earliest, *B. diversus* workers appeared the latest, and the appearance of *B. hypocrita* workers was intermediate between them (Fig. 5).

Except for *B. ardens*, models with only intercepts were fitted to the HW' of trapped workers (Table 2). In *B. ardens*, the date had a significantly positive parameter, indicating a seasonal increase in the worker HW' (Fig. 5c). The urbanization also had a significantly positive parameter in *B. ardens*.

Morphology

Variation in the seven morphological characters among 96 females of the four abundant species was summarized into the first and second principal components that contributed to 82 and 16%, respectively, of the total variation. The first principal component represented the overall body size, and the second one reflected the tongue length (PL and GL) compared to the HW, MW, and CL (Fig. 6a). Morphological variation among the four species was summarized into the first (83%) and second

Table 2 Parameter estimates in generalized linear models for the head width (HW) of trapped workers of four abundant species

Parameter	Estimate in selected model for the worker head width (mm) of			
	<i>Bombus terrestris</i>	<i>B. hypocrita</i>	<i>B. ardens</i>	<i>B. diversus</i>
Intercept	4.010***	4.041***	3.546***	3.641***
Date ^a	NS	NS	0.090***	NS
Land use				
Urbanization (<i>p</i> 1)	NS	NS	0.054**	NS
Paddy field prevalence (<i>p</i> 2)	NS	NS	NS	NS
Trap height (<i>h</i>)	NS	NS	NS	NS
Local abundance				
<i>B. terrestris</i> (<i>e</i>)	NS	NS	NS	NS
Other native species (<i>n</i>)	NS	NS	NS	NS

NS parameters are not selected in the generalized linear model with the lowest BIC (any interactions were not selected)

* $0.01 \leq P < 0.05$; ** $0.001 \leq P < 0.01$; *** $P < 0.001$ (Wald's *t* statistics: parameters different from zero)

^aThe number of dates after January 2004

(11%) canonical discriminant variables. The first discriminant variable represented a contrast between the tongue length (PL and GL) and the CL, and the second one represented the ML relative to the HW (Fig. 6b).

Although females showed large variation in body size within species, body shape clearly discriminated among the species. The three native species had short (*B. hypocrita*), intermediate (*B. ardens*), and long (*B. diversus*) tongues (PL + GL) compared with the HW (Fig. 6c). The relative tongue length [(PL + GL)/HW] of *B. terrestris* was intermediate between *B. hypocrita* and *B. ardens* (Fig. 6c). The relative wing length (ML/HW) was the longest in *B. diversus* and was the shortest in *B. terrestris* (Fig. 6d).

Discussion

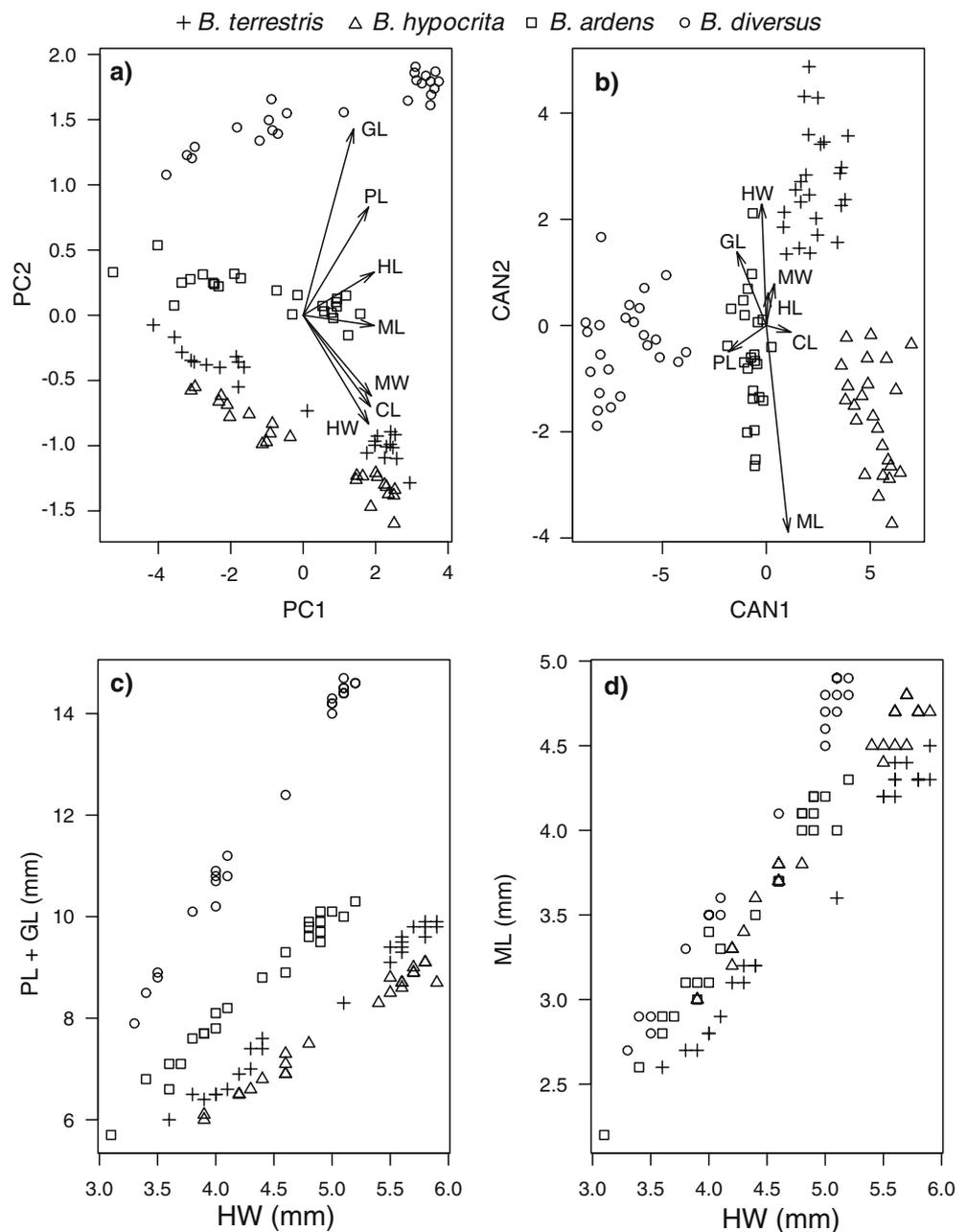
Bumblebees have been the object of numerous studies on competition for food (Goulson 2003a). Field experiments manipulating competitors are necessary to show competitive effects on foraging efficiency, reproductive success, and population growth (Bowers 1985b; Thomson 2004). Alternatively, field observations are also effective to discover potential competitive interactions (Forup and Memmott 2005). Patterns of negative associations between species in abundance, body size, and other measurements for population and fitness suggest interspecific competition. However, such patterns can result from factors other than competition, such as different habitat preference in heterogeneous environments. Thus, the patterns observed in the fields should be examined when considering habitat types, environmental conditions, and other factors related with the population and fitness of bumblebees (Goulson et al. 2002).

In this study, we considered land use and some related factors when we examined the abundance and body size of exotic and native bumblebees. For example, the spatial patterns of trapped bumblebees indicated a negative association in abundance between exotic

B. terrestris and native *B. ardens* in our previous study carried out at eight trapping sites on a transect (Inari et al. 2005). In the present study conducted at more sites, the abundance of *B. ardens* was not the lowest at the sites where *B. terrestris* was the most abundant although the abundances of the two species were negatively associated (Fig. 4). Statistical models in the present study suggest that the different habitat preferences between the exotic and native species result in the negative association in their abundances (Table 1). When land use was taken into account in the models, the local abundance of *B. terrestris* was not associated negatively with the number and body size of native bumblebees (Tables 1, 2). Thus, we did not find any competitive interactions from *B. terrestris* to the native bumblebees although habitat conditions seem to be major determinants of the native bumblebee populations.

The exotic and native species showed different trends with regard to the effects of land use on the number of trapped bumblebees. *B. terrestris* were more frequently trapped in less-urbanized sites where paddy fields were prevalent, and this habitat preference intensified when the native species were abundant (Table 1). On the other hand, the native species showed the opposite trend in their habitat preference. Successful invasions of *B. terrestris* into the lowland with many paddy fields were found also in Mukawa, Hidaka district, Hokkaido (Matsumura et al. 2004). It is not clear whether the examined species prefer different habitats because these species share subterranean nest sites. Among them, *B. terrestris*, *B. hypocrita*, and *B. diversus* prefer similar nest sites, such as ridges at the edges of fields and banks along irrigation channels (Nakajima et al. 2004). In Europe, nest site preference differs among species with regard to habitat types on the margins of fields and forests (Svensson et al. 2000; Kells and Goulson 2003). Thus, there is a possibility that the native species have habitat preference that is different from that of *B. terrestris*. Furthermore, urbanization was positively associated with abundance of *B. ardens* (Table 1). In the urban district of Tokyo, *B. ardens* made its nests in the

Fig. 6 Morphology of females of *B. terrestris* (crosses), *B. hypocrita* (triangles), *B. ardens* (squares), and *B. diversus* (circles). **a** Principal component analysis. Arrows show the vectors of the loading factors for the *PC1* and *PC2* in seven characters shown in Fig. 2. **b** Canonical discriminant analysis to discriminate the four species. Arrows show the vectors of the standardized coefficients of the first and second canonical components (*CAN1* and *CAN2*) in the seven characters. **c** Relationship between HW (mm) and total length of PL and GL (mm). **d** Relationship between HW (mm) and ML (mm)



covered place above the ground surface, such as wooden houses and nests of birds (Kuboki and Ochiai 1985). This nesting ability of *B. ardens* may be responsible for its abundance in urban habitats.

The body size of *B. ardens* workers varied temporally and spatially (Table 2), probably due to different availability of food. As shown in previous studies (Harder 1985; Inoue and Kato 1992), the worker HW' increased seasonally, which may reflect the increase in pollen harvest as worker populations of colonies increase. The positive effects of urbanization on body size of *B. ardens* are difficult to interpret.

Commercial colonies of *B. terrestris* used in greenhouses are the sources of its populations in the wild. In

our previous study, we found that the number of commercial colonies used within 4-km radii around traps was positively correlated with the number of trapped *B. terrestris* (Inari et al. 2005). In this study, we applied a two-dimensional diffusion function to the spatial distribution of trapped *B. terrestris* and estimated a dispersal curve from commercial colonies in greenhouses. The curve indicated that 99% of *B. terrestris* individuals that had originated from fugitives from greenhouses had dispersed within a 18.8-km radius by 2004 since the utilization of its commercial colonies began in 1992. *B. terrestris* had spread about 85 km south, 55 km west, 20 km east, and 65 km north by 1999 in Tasmania, where it was first observed in 1992 (Stout and Goulson 2000). The invasion

rate seems to be slower in Hokkaido than in Tasmania. However, it is difficult to compare the rates of spread between the two regions because the methods to measure the abundance of *B. terrestris* were different.

The number of trapped bumblebees of two native species, *B. hypocrita* and *B. diversus*, was positively associated with the local abundance of *B. terrestris* (Table 1). This result disagrees with the pattern expected from competition between exotic and native bumblebees. These positive associations are likely to result from variation in trapping efficiency or carrying capacity among trapping sites. The efficiency of catches using window traps may be sensitive to local environmental conditions, such as the wind speed, open space distribution, and flowering events around the traps. Carrying capacity of local populations may differ depending on available resources: nectar, pollen, and nest sites (Bowers 1985a, 1986). If the environmental conditions or available resources vary among the trapping sites, many bumblebees are caught at sites where trapping is efficient or resources are rich; however, a few are caught at ineffective or poor sites, which results in the positive associations of catches among species. Because it is difficult to measure both trapping efficiency and carrying capacity directly, these factors can hardly be included in

the analysis on observed patterns. Therefore, field experiments in which *B. terrestris* density is manipulated are necessary to show competition between exotic and native bumblebees. Native species that share resources with *B. terrestris* should be examined in the experiments.

The morphology of the mouth, in particular the tongue length, is closely related with the utilization of floral resources (Harder 1985; Inoue and Kato 1992). The morphology of *B. terrestris* females was intermediate between *B. hypocrita* and *B. ardens* females, and variation in the tongue length considerably overlapped among the three species. This result suggests that *B. terrestris* shares floral resources with both *B. hypocrita* and *B. ardens*, which is also suggested by records of flower visitation in Hokkaido (Matsumura et al. 2004). Thus, not only *B. hypocrita* but also *B. ardens* should be examined in the future experiments.

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Appendix 1 Locations and altitudes of 17 trapping sites, land use patterns within 1.5-km radii around the sites, and heights of 70 traps at the sites

No.	Site	Latitude (42°N)	Longitude (141°E)	Altitude (m)	Land use (%)				Trap height (m)
					Paddy field	Farm	Urban area	Woodland	
1	Minaminagamuna-nishi	57.1'	39.2'	7	55	32	0	1	11.4, 12.2, 1.3, 1.5
2	Hoei	56.9'	36.0'	8	38	51	1	5	8.4, 7.0, 1.6, 1.2
3	Daigakubashi	54.5'	40.3'	8	33	46	1	5	7.8, 9.4, 1.4, 1.4
4	Minaminagamuna-higashi	57.6'	41.3'	9	33	61	1	2	11.3, 11.8, 1.7, 1.5
5	Syukubai-kita	52.7'	42.0'	9	0	84	1	9	9.5, 8.8, 1.1, 0.9
6	Kamiyamaguchi-kita	54.5'	37.6'	10	38	53	1	6	10.5, 10.2, 1.4, 1.6
7	Osatsu	53.2'	38.6'	11	12	63	1	6	12.4, 10.1, 1.6, 1.4
8	Neshikoshi	51.3'	39.7'	11	0	54	10	14	10.1, 12.6, 11.3, 1.1, 1.2, 1.2
9	Chitose	50.0'	39.6'	12	0	4	78	3	11.3, 13.3, 1.4, 1.7
10	Syukubai-higashi	51.5'	42.4'	13	0	36	1	34	9.0, 9.0, 1.5, 1.2
11	Eniwa-kita	53.5'	36.1'	21	5	48	29	8	8.7, 12.2, 1.4, 1.6
12	Shimamatsu-kita	55.7'	34.5'	22	5	56	27	4	10.0, 9.4, 1.5, 1.3
13	Osatsu-eki	51.3'	37.3'	22	0	41	42	13	7.7, 9.3, 1.7, 1.1
14	Horonai	56.4'	43.6'	29	5	68	1	19	10.2, 9.5, 1.2, 1.1
15	Eniwa-higashi	51.9'	35.8'	32	0	34	38	15	11.3, 10.2, 1.3, 0.9
16	Izumisato	54.3'	43.7'	43	9	48	2	31	10.4, 6.8, 1.2, 1.1
17	Shimamatsu-nishi	54.7'	32.9'	56	0	20	16	26	9.6, 10.9, 1.4, 1.1

Appendix 2 Mean number of bumblebees collected from traps and mean HW' of workers of six species trapped at 17 sites

Site	Mean number of trapped bumblebees of							Mean HW' (mm) of trapped workers of					
	<i>ter</i>	<i>hyp</i>	<i>ard</i>	<i>div</i>	<i>pse</i>	<i>hpn</i>	Total	<i>ter</i>	<i>hyp</i>	<i>ard</i>	<i>div</i>	<i>pse</i>	<i>hpn</i>
Minaminagamuna-nishi	5.25	0.00	0.00	0.00	0.50	0.00	5.75	3.95					3.57
Hoei	14.75	6.75	0.75	0.75	0.00	0.00	23.00	3.98	4.02	3.21			
Daigakubashi	7.75	0.50	0.00	0.50	0.00	0.00	8.75	4.05	4.00		3.54		
Minaminagamuna-higashi	3.00	0.00	0.00	2.00	0.50	0.00	5.50	4.02			3.59		
Syukubai-kita	2.75	0.50	0.75	1.25	0.50	0.00	5.75	4.28		3.77			
Kamiyamaguchi-kita	23.00	1.50	1.75	8.25	0.00	0.00	34.50	3.98	4.14	3.25	3.60		
Osatsu	7.25	0.75	0.75	1.00	0.75	0.00	10.50	3.95	4.17		3.88	3.45	
Neshikoshi	3.67	1.83	10.33	2.00	0.67	0.00	18.50	4.08	3.99	3.52	3.93	3.65	
Chitose	0.25	0.50	21.00	0.25	0.00	0.00	22.00		4.23	3.61			
Syukubai-higashi	0.75	0.25	1.50	3.25	0.25	0.00	6.00	4.37		3.74	3.74	3.65	
Eniwa-kita	5.25	0.50	0.50	0.00	0.00	0.00	6.25	4.10		3.60			
Shimamatsu-kita	4.75	4.00	7.00	4.50	0.00	0.00	20.25	4.05	4.04	3.48	3.66		
Osatsu-eki	1.00	2.00	4.50	2.50	0.25	0.00	10.25	3.97	4.08	3.56	3.80		
Horonai	3.00	3.50	0.25	6.75	0.00	0.00	13.50	3.86	4.14		3.54		
Eniwa-higashi	1.75	2.00	10.75	1.25	0.00	0.00	15.75	4.17	4.09	3.52	3.54		
Izumisato	0.75	1.00	8.25	1.50	0.25	0.25	12.00	3.87	3.20	3.49	3.65		3.86
Shimamatsu-nishi	1.50	1.00	0.50	0.00	0.00	0.00	3.00	3.77	3.79				
Mean	5.08	1.56	4.03	2.10	0.22	0.01	13.01	4.03	3.99	3.52	3.68	3.58	3.86

ter: *B. terrestris*, *hyp*: *B. hypocrita*, *ard*: *B. ardens*, *div*: *B. diversus*, *pse*: *B. pseudobaicalensis*, *hpn*: *B. hypnorum*

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