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ORIGINAL PAPER

An assessment of the habitats and trophic level of the introduced slug *Limax maximus* in Japan

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Abstract *Limax maximus* is one of the most widely introduced terrestrial gastropods in the world. The first record of the species in Japan was published in 2009, and its distribution range has rapidly expanded. Our aims were to estimate the habitat characteristics and diet of introduced *L. maximus* in Japan. We conducted field surveys at 21 sites in Ibaraki Prefecture, where *L. maximus* is frequently reported by citizens. We found that the habitat range of *L. maximus* is broad, occurring from rural to urbanized areas; this is distinctively different from the case of native slugs and non-native slugs previously introduced to Japan. To characterize *L. maximus* diets, we performed stable isotope analyses on individuals and the species' potential food sources. *Limax maximus* consistently showed high δ^{13}C and δ^{15}N, meaning that it occupies a similar trophic position as predatory invertebrates such as spiders and ground beetles. Analyses of the stable isotope data by using a Bayesian mixing model (MixSIAR) suggested that *L. maximus* feeds on various food sources but mainly mushrooms. Our findings imply that *L. maximus* is a generalist with regard to habitat preference and has a flexible diet; these are likely factors in its rapid population increase. Currently, the Japanese government designates *L. maximus* as an introduced species that requires comprehensive countermeasures. We recommend sharing the species' occurrence information with the public and cautioning farmers to be aware of this slug and its detrimental effects, so that they may take measures against its potential agricultural damage.

Keywords Diversity · Diet · Habitat preference · Introduced species · Leopard slug · Terrestrial gastropod · Stable isotope

Introduction

Terrestrial gastropods have invaded many parts of the world (Cowie and Robinson 2003; Gerlach et al. 2021; Mito and Uesugi 2004; Sessions and Kelly 2002). Predation, competition, and behavioral interference

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are considered to be major threats posed by invasive terrestrial gastropods, as they feed on native plants and animals (Coote and Loève 2003; Gerlach et al. 2021; Hahn et al. 2011; Joe and Daehler 2008), show overlap in the food sources of native species (Paustian and Barbosa 2011), exhibit aggression and attacking behavior (Kimura and Chiba 2010), and have chemical influences through their mucus trails that is disliked by certain species (O'Hanlon et al. 2020). At the community level, these effects can alter species composition and reduce diversity (Aubry et al. 2005; Horsák et al. 2013; Tomiyama 2002). Many locally endemic species of terrestrial gastropods have limited migration ability. Consequently, invasive terrestrial gastropods can modify native species' composition, making the management of invasive species an urgent issue.

The Japanese archipelago is characterized by rich native terrestrial-gastropod communities (Azuma 1982), but it has faced frequent invasion by non-native slugs. In the early Meiji period (1868–1912), *Limacus flavus* (Linnaeus 1758)—native to Europe—was introduced and caused serious damage to agriculture in Japan (Takashima 1954). After World War II, the European slug *Ambigolimax valentianus* (A. Férussac, 1821) was introduced, and *L. flavus* disappeared from most areas of Japan (Kurozumi 2002). *Ambigolimax valentianus* is now one of the most dominant slugs in Japan, causing agricultural damage in many areas. In 2006, a large slug native to Europe, the leopard slug *Limax maximus* Linnaeus, 1758, was discovered in Ibaraki Prefecture (Hasegawa et al. 2009), and its distribution is rapidly increasing (Iijima et al. 2013; Morii et al. 2015). *Limax maximus* has also been introduced to North America, South America, Africa, and Australia (Barker 2015; Barker and McGhie 1984; Roth and Sadeghian 2006). In the continental United States and Europe, it is seen as an agricultural pest (Kozłowski 2012; Stange and Deisler 2005). The Ministry of the Environment of Japan has listed *L. maximus* as an invasive alien species that requires the application of comprehensive measures to avoid damage to biodiversity, the economy, and industries. The Plant Protection Act of Japan prohibits the import of *L. maximus*.

Unfortunately, *L. maximus* is known to exhibit aggressive behavior toward other slugs (Rollo and Wellington 1979) and may feed on the eggs of native species (Watz et al. 2022). Such behavior may cause serious damage to native communities at sites where *L. maximus* is introduced (hereafter referred to as introduced sites). However, little information is available on its ecological impacts. *Limax maximus* is considered omnivorous (Taylor 1902), feeding on both plants (Joe 2006; Komatsu and Saeki 2022) and animals (Norden and Williams 2015). On the other hand, the species appears to often feed on fungi (Keller and Snell 2002). In Hawaii, where this species has also been introduced, stable isotope analysis indicates that animals may be its primary food resource (Meyer and Yeung 2011). Therefore, the results from previous reports are not consistent, and traits may change in different areas where it has become established. Our objective was to examine the ecological effects of the introduction of *L. maximus* in Japan by exploring its habitat characteristics, differences in species composition of terrestrial gastropods between sites where it is present and absent, and its feeding habits.

Materials and methods

Habitat characteristics, species composition, and diversity

To identify the habitat characteristics of *L. maximus*, we performed a field survey at 21 localities in southern Ibaraki Prefecture from July 2021 to November 2022 (Fig. 1; Supplementary Material 1). This region includes the place where the species was first reported in Japan (Hasegawa et al. 2009). Since then, there have been frequent reports from the public about the species' occurrence to the Ibaraki Nature Museum. In selecting survey sites, we referred to previous reports by members of the public to the museum, information in the Social Networking System on websites (searching for the terms "*Limax maximus*" and "madarakouranamekuji," the common name in Japanese), and information in the literature (e.g., Hasegawa et al. 2009; Namba et al. 2020) to ensure that an adequate number of *L. maximus* presence sites (10) for analysis were included. In addition, we arbitrarily selected sites near the presence sites (i.e., within a 10-km radius) with similar geographical features (elevation, topography, and geological history) to make the number of presence and absence sites generally equal.

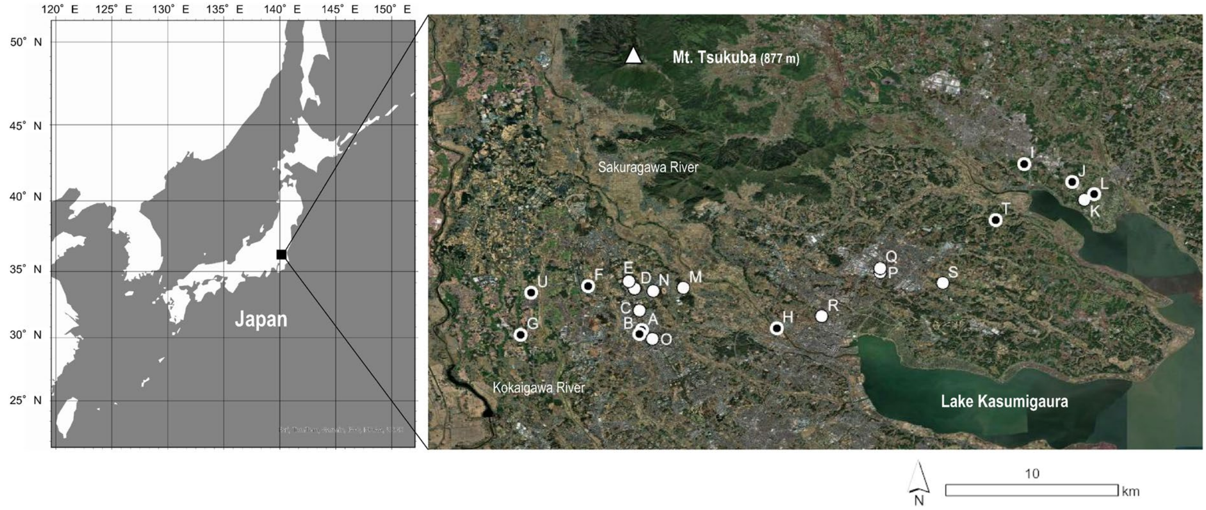


Fig. 1 Map of Japan (left) and inset of southern Ibaraki Prefecture showing the field survey sites (right). Black and white circles indicate sites where *Limax maximus* was recorded or not recorded, respectively. See Supplementary Material 1 for details of the field survey sites

The 21 sites covered a wide environmental gradient from urban to rural areas. At these sites, one or two investigators looked for *L. maximus* and any other slugs and land snails with a shell size over 5 mm in diameter to maximize the sampling efficiency. Survey efforts were set to 2 h per person, and the area for investigation was set to 1 ha. Species names and the number of individuals, including dead shells, were recorded at each site. Furthermore, we conducted a beer-trap survey. Some slugs, including *L. maximus*, are attracted to the flavor of beer (Udaka and Tanaka 2010; Komatsu, personal observation). At each study site, we set three traps (7 cm diameter and 5 cm height) with beer on the ground for 48 h, and the results were used to confirm the absence of individuals at a site during the field survey.

For each of the 21 sites, we classified land-use type using ArcGIS ver. 10.4.1 (ESRI). Using a vegetation map (1/25,000 scale; Ministry of Environment, Japan), we classified the land use within each 1-ha survey site into three types: urban area, farmland, or forest. Urban area included residential and industrial areas; farmland included rice paddy, vegetable fields, grassland, and fruit gardens; and forest included lands dominated by oaks, chestnut, willow, bamboo, or other tall trees. The vegetation type was also confirmed by field observation.

Cluster analyses were performed on the basis of the proportion of the three land-use types (i.e., urban area, farmland, and forest), and we examined the relationships among the occurrence of *L. maximus*, habitat characteristics, and species composition of terrestrial gastropods. The analyses were conducted by using R 4.4.1 (R Core Team 2022) with the *vegan* package (Oksanen et al. 2022). We used the Horn-Morisita index (Krebs 1999) as a distance measure to quantify the similarity of the land-use types.

To characterize the communities of terrestrial gastropods at sites where *L. maximus* had been introduced, alpha and beta diversity were quantified by using the data obtained at the 21 sites. For the analyses, we used the total number of living and dead shells as abundance data. We determined species richness (S) and Shannon–Wiener (H') indices as indicators of alpha diversity and compared their means between *L. maximus* presence ($n = 10$) and absence ($n = 11$) sites. When we performed the analyses, we used a data set that excluded *L. maximus*. Student's t-tests were used to test differences in alpha diversity (S and H'). For beta diversity, we performed principal coordinate analyses (PCoA) by using the species' composition data. In this process, we divided the data set into two groups depending on the presence and absence of *L. maximus*. Then, the mean distance of each site from the centroid was compared between the two groups. We hypothesized that, if beta diversity differed between the two groups, their mean distances to the centroid would be significantly different. We performed this analysis by using presence–absence (i.e., qualitative) and abundance (i.e., quantitative) data. Distance was calculated by using the Horn-Morisita index (Krebs 1999). To compare species composition between the two types of study site (i.e., with and without *L. maximus*), we performed non-metric multidimensional scaling (NMDS) analyses with qualitative and quantitative data. Permutation multivariate analyses of variance (PERMANOVA) were run to test for statistical differences.

Feeding habits

Stable isotope analyses were conducted to measure stable isotope ratios in the bodies of prey and predators, and the values were compared to estimate what kind of food the predator ate and to what extent the food was assimilated into the body (McCue et al. 2020). Samples were collected from May to November in 2022 at two sites where we conducted field surveys (Fig. 1). The first site is part of the Kasuga campus of the University of Tsukuba (site A in Fig. 1), which is highly urbanized. The other site is at Kitanoiri, in the city of Ishioka (site I in Fig. 1). This site is located in a rural *satoyama* area (Takeuchi 2001) covered by forest, rice paddy, and grassland, and the landowner grows mushrooms as well. We selected these contrasting sites to examine whether the diet of *L. maximus* varied with the land-use type. We collected 12 individuals of *L. maximus* per site (Supplementary Material 2). For reference, we also collected sympatrically occurring species of terrestrial gastropods, including two slug species, *A. valentianus* (non-native) and *Meghimatium bilineatum* (W. H. Benson, 1842) (native). As an indicator of trophic positions, earthworms, millipedes, and pillbugs (decomposers); grasshoppers and scarab beetles (herbivores); and spiders and ground beetles (carnivores) were also collected. As candidate food resources, we collected vascular plants, leaf litter, bryophytes, aerial algae (i.e., algae creating terrestrial biofilms, often found on tree trunks, soil, and rocks), lichens, mushrooms, and animals (see Supplementary Material 2). The number of samples per species or taxonomic group ranged from 1 to 12 per site.

Collected samples were brought back to the laboratory and stored in a freezer at − 20 °C. The samples were then dried at 60 °C for 48 h. We used snail foot muscle for the analyses of terrestrial gastropods, in accordance with the method of Němec et al. (2021). After being dried, the tissues were ground and each sample was stored in a glass vial and weighed. Samples were analyzed by using a mass spectrometer (DELTA V Advantage, Thermo Fisher Scientific Inc., Waltham, MA, USA) at the Center for Ecological Research, Kyoto University, Japan, in accordance with the protocol developed by the center. Stable isotope ratios of carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) were determined as C and N relative to Pee Dee Belemnite limestone and atmospheric N, respectively, by using the following equation: $\delta^{13}C$ or $\delta^{15}N = (R_{sample}/R_{standard} - 1) \times 1000$, in which R_{sample} is the stable isotopic ratio in the samples, and $R_{standard}$ is the ratio in the standard.

To estimate the proportions of food resources of *L. maximus*, we used MixSIAR (Stock et al. 2018) to create and run the Bayesian mixing models. In the analyses, the mean trophic fractionations of $\delta^{15}N$ and $\delta^{13}C$ were set at 3.4‰ (± 1.0‰) and 0.4‰ (± 1.3‰), respectively (Post 2002), along with the stable isotope data of *L. maximus* as a consumer. The sample size of *L. maximus* was 12 at each site, and we ran the analyses for each site. For comparison, we performed the analyses for two other slug species, *M. bilineatum* (native) and *A. valentianus* (non-native) collected at the urban site. The sample size was 12 for these species as well. We used the stable isotope ratios of aerial algae, bryophytes, lichens, vascular plants, animals, and mushrooms, as candidate food resources of consumers (i.e., *L. maximus* and the two other slug species). By using these data, we estimated the proportion of diets as posterior probability distributions by running Markov chain Monte Carlo (MCMC) simulation in the Bayesian tracer (i.e., stable isotope) mixing model framework (Stock et al. 2018). MCMC was considered to have converged when the Gelman–Rubin statistic (Gelman and Rubin 1992) was less than 1.1.

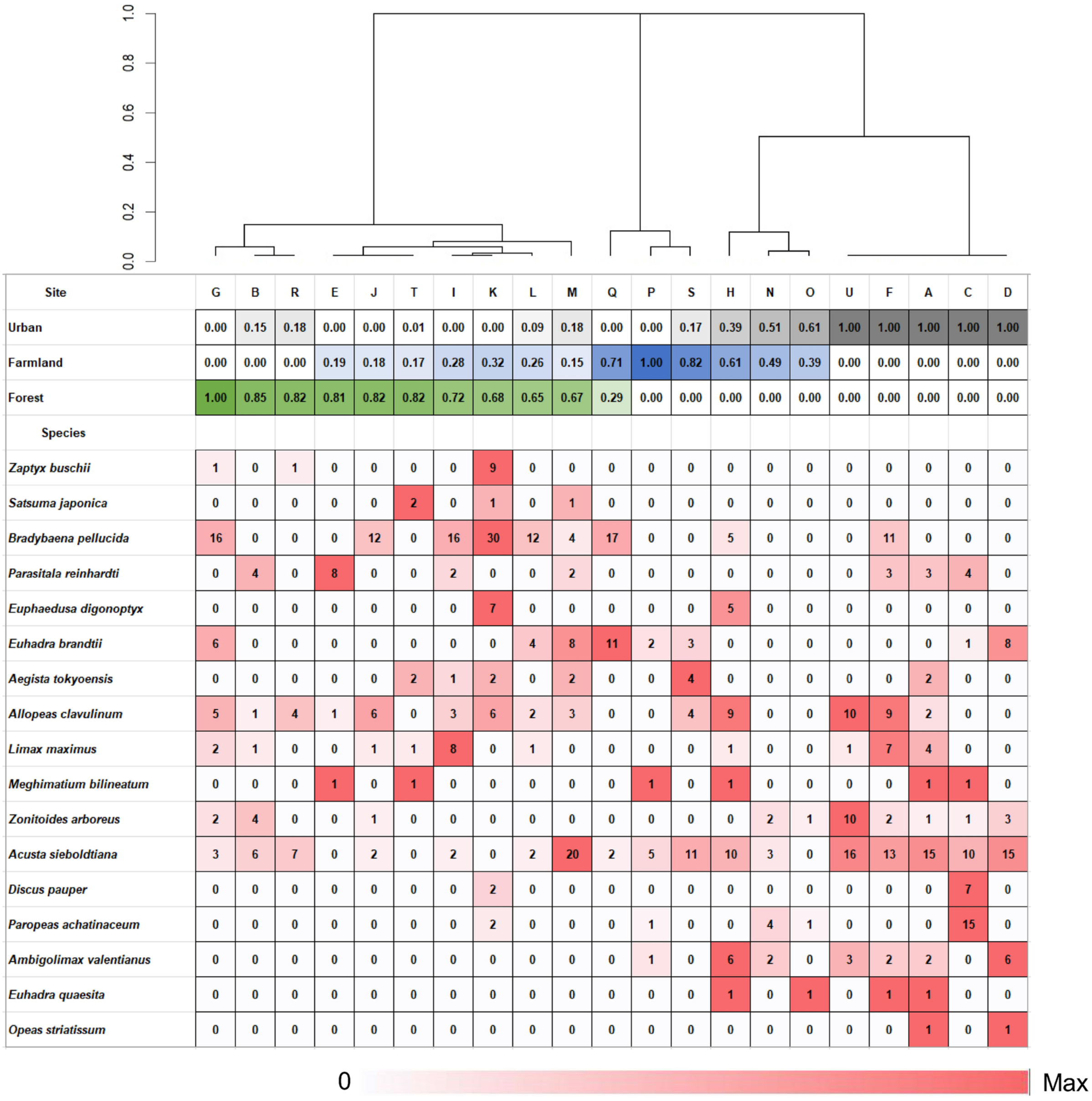
Ethical note

All the experiments and collections were performed in accordance with the Institutional Policy on Animal Experimentation of the University of Tsukuba.

Results

Habitat characteristics, species composition, and diversity

In the ground surveys, we recorded a total of 20 species of terrestrial gastropods, including 578 dead



Site	G	B	R	E	J	T	I	K	L	M	Q	P	S	H	N	O	U	F	A	C	D
Urban	0.00	0.15	0.18	0.00	0.00	0.01	0.00	0.00	0.09	0.18	0.00	0.00	0.17	0.39	0.51	0.61	1.00	1.00	1.00	1.00	1.00
Farmland	0.00	0.00	0.00	0.19	0.18	0.17	0.28	0.32	0.26	0.15	0.71	1.00	0.82	0.61	0.49	0.39	0.00	0.00	0.00	0.00	0.00
Forest	1.00	0.85	0.82	0.81	0.82	0.82	0.72	0.68	0.65	0.67	0.29	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Species																					
Zaptyx buschii	1	0	1	0	0	0	0	9	0	0	0	0	0	0	0	0	0	0	0	0	0
Satsuma japonica	0	0	0	0	0	2	0	1	0	1	0	0	0	0	0	0	0	0	0	0	0
Bradybaena pellucida	16	0	0	0	12	0	16	30	12	4	17	0	0	5	0	0	0	11	0	0	0
Parasitala reinhardti	0	4	0	8	0	0	2	0	0	2	0	0	0	0	0	0	0	3	3	4	0
Euphaedusa digonoptyx	0	0	0	0	0	0	0	7	0	0	0	0	0	5	0	0	0	0	0	0	0
Euhadra brandtii	6	0	0	0	0	0	0	0	4	8	11	2	3	0	0	0	0	0	0	1	8
Aegista tokyoensis	0	0	0	0	0	2	1	2	0	2	0	0	4	0	0	0	0	0	2	0	0
Allopeas clavulinum	5	1	4	1	6	0	3	6	2	3	0	0	4	9	0	0	10	9	2	0	0
Limax maximus	2	1	0	0	1	1	8	0	1	0	0	0	0	1	0	0	1	7	4	0	0
Meghimatium bilineatum	0	0	0	1	0	1	0	0	0	0	0	1	0	1	0	0	0	0	1	1	0
Zonitoides arboreus	2	4	0	0	1	0	0	0	0	0	0	0	0	0	2	1	10	2	1	1	3
Acusta sieboldtiana	3	6	7	0	2	0	2	0	2	20	2	5	11	10	3	0	16	13	15	10	15
Discus pauper	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	7	0
Paropeas achatinaceum	0	0	0	0	0	0	0	2	0	0	0	1	0	0	4	1	0	0	0	15	0
Ambigolimax valentianus	0	0	0	0	0	0	0	0	0	0	0	1	0	6	2	0	3	2	2	0	6
Euhadra quaesita	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	1	0	1	1	0	0
Opeas striatissum	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	1

Fig. 2 Results of cluster analyses based on the proportion of land cover around the survey sites and the corresponding occurrences of terrestrial gastropods found in the field surveys. Intensity of gray, blue, and green shading indicates the proportion of landcover. Intensity of the red shading indicates the numbers of individuals recorded in the survey. Species recorded at more than one site are shown. See Supplementary Material 3 for the full occurrence data

and living individuals (Supplementary Material 3). According to the cluster analyses, the 21 study sites were aligned by an urbanization gradient (Fig. 2). The least urbanized site was G, with all the area covered by forest, whereas sites A, C, D, F, and U were fully dominated by urban areas. The rest of the sites were a mixture of two or three land-use types. *Limax maximus* occurred at 10 of the 21 sites, with a broad habitat range covering forest, farmland, and urbanized areas (Fig. 2). The proportions of forest and urban area were each not significantly different between sites with and without *L. maximus* (*t*-test, $p = 0.52$ for forest and $p = 0.40$ for urban area). The other slugs we recorded were *A. valentianus*, *M. bilineatum*, and *Meghimatium fruhstorferi* (Collinge, 1901). Five of the 20 recorded species had been introduced from other countries or other regions of Japan. Non-native *Zonitoides arboreus* (Say, 1817), like *L. maximus*, occurred in both forested and urban areas. On the other hand, non-native *Paropeas achatinaceum* (L. Pfeiffer, 1846) and *A. valentianus* occurred exclusively in urban areas. *Bradybaena pellucida* Kuroda & T. Habe, 1953, originating from the western part of Japan, occurred mainly from forested to farmland areas. The habitats of native species varied widely among the species. The most common species occurring at numerous sites was *Acusta sieboldtiana* (L. Pfeiffer, 1850), followed by *Allopeas clavulinum kyotoense* (Pilsbry & Y. Hirase, 1904). The other native species were recorded at a relatively limited number of sites (i.e., two to seven). The species that were dominantly recorded at rural sites were *Zaptyx buschii* (Küster, 1844) and *Satsuma japonica* (L. Pfeiffer, 1847). In contrast, the species exclusively recorded in urbanized areas were *Euhadra quaesita* (Deshayes 1850) and *Opeas striatissum* (Gredler 1882). The absence of *L. maximus* at the 11 sites was confirmed by the beer-trap surveys as well.

In a diversity analysis of species other than *L. maximus*, the average species richness was 5.4 at sites where *L. maximus* was recorded and 4.9 at sites where it was not recorded (Fig. 3a; $t = 0.58$, df = 19, $p = 0.47$). The Shannon–Wiener index (H') was 1.34 and 1.24 at sites with and without *L. maximus*, respectively (Fig. 3b; $t = 0.91$, df = 19, $p = 0.37$). According to the PCoA using the presence–absence data, the average distance from each study site where *L. maximus* occurred to the centroid of coordinates was 0.34. This was lower than the average distance between each of the sites where *L. maximus* did not occur and the centroid of coordinates (0.47, $p = 0.05$ by permutation test, $n = 999$; Fig. 3c). Application of the quantitative data indicated that the distance from each site to the centroid of coordinates at presence sites (0.40) was smaller than that at absence sites (0.46), yet the difference was not statistically significant ($p = 0.48$; Fig. 3d). According to the PERMANOVA, there was no difference in species composition between sites where *L. maximus* was recorded and not recorded ($p = 0.51$ for qualitative and $p = 0.40$ for quantitative data sets). The ordination diagrams based on NMDS analyses indicated that the sites where *L. maximus* was not recorded overlapped with those where *L. maximus* was recorded and were more widely dispersed in the ordination diagrams (Fig. 4).

Feeding habits

According to the stable isotope analyses conducted at an urban and a rural site, *L. maximus* had markedly higher δ^{13}C and δ^{15}N values than did vascular plants, aerial algae, and bryophytes (Fig. 5). At the urban site, the highest δ^{15}N was recorded in mushrooms (3.58‰ ± 3.63‰), followed by cicadas (3.35‰ ± 2.18‰). The δ^{15}N (2.55‰ ± 1.31‰) of *L. maximus* was similar to those of predators (e.g., spiders and ground beetles), decomposers (e.g., carrion beetle), and herbivore (e.g., scarab beetle; Fig. 5a). The other introduced slug, *A valentianus*, had a δ^{15}N value similar to that of *L. maximus*, but the land snails, such as *A. sieboldtiana* (0.23‰ ± 1.38‰), *M. bilineatum* (−4.75‰ ± 2.58‰), and *Euhadra brandtii brandtii* (Kobelt 1875) (−5.57‰ ± 1.46‰), had lower values.

At the rural site, δ^{15}N of *L. maximus* (5.24‰ ± 1.18‰) was also high; its ratio was similar to that of carnivorous carabid beetles (4.77‰ ± 2.84‰) and higher than that of *A. valentianus* (3.26‰ ± 0.32‰; Fig. 5b). δ^{13}C of *L. maximus* (−26.3‰ ± 0.60‰) was estimated to be similar to that of *A. valentianus* (−26.4‰ ± 0.07), which was between the values of earthworms (−27.8‰ ± 0.40‰) and carrion beetles (−25.0‰ ± 0.66‰). For mushrooms, the values of δ^{15}N (3.78‰ ± 1.17‰) and δ^{13}C (−26.6‰ ± 1.16‰) were close to those of *L. maximus*.

According to Bayesian mixing analysis using MixSIAR, *L. maximus* collected at the urban site

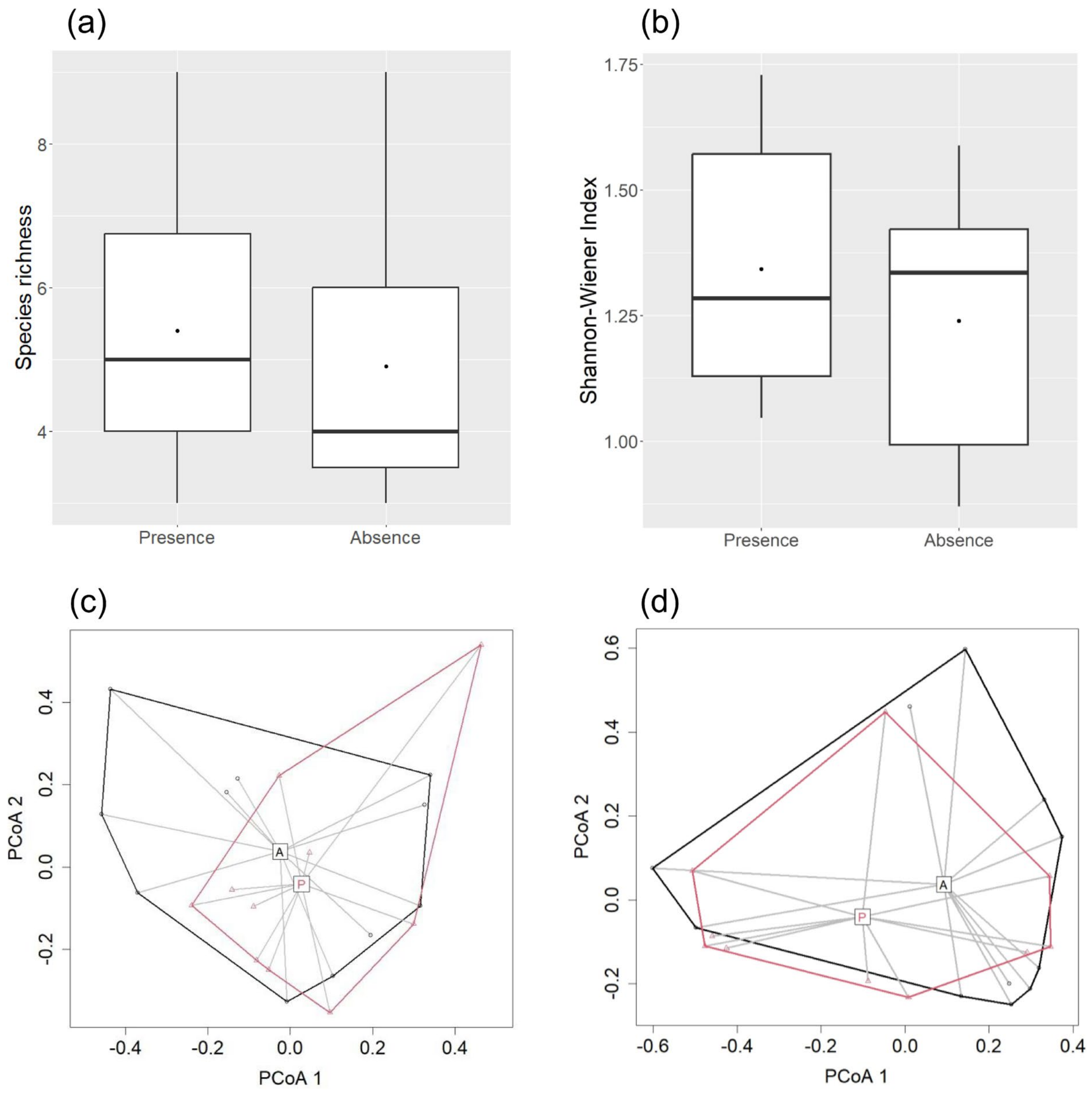


Fig. 3 Comparisons of species diversity between sites where *Limax maximus* was recorded (i.e., present) or not recorded (absent) excluding the data on *L. maximus*: **a** species richness, **b** Shannon–Wiener index, **c** multivariate dispersion for the groups of sites where *L. maximus* was present (P, red) and absent (A, black), as determined from presence–absence data, **d** multivariate dispersion for the groups of sites where *L. maximus* was present and absent, as determined from quantitative data. Species richness and the Shannon–Wiener index were used as indicators of alpha diversity whereas multivariate dispersions were used as indicators of beta diversity

was estimated to feed on mushrooms as the highest proportion, followed by animals (Table 1; Fig. 6a). An even higher proportion of mushrooms was found in the samples from rural site (Fig. 6b). In contrast, the highest proportion of the diets of *M. bilineatum* collected at the urban site was estimated to be lichens, followed by bryophytes and aerial algae (Fig. 6c). *Ambigolimax valentianus* collected at the

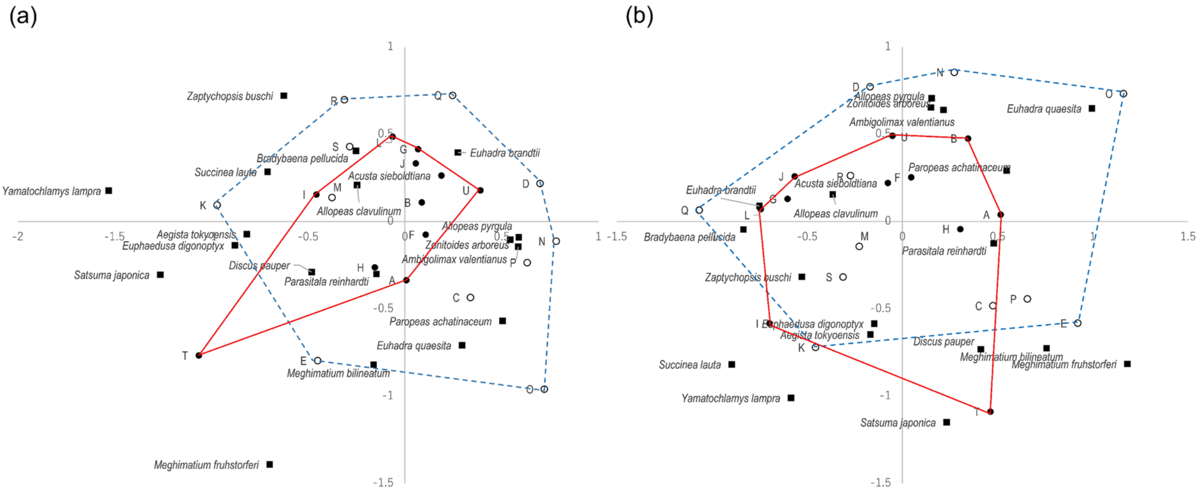


Fig. 4 Ordination analyses (non-metric multidimensional scaling) of species composition at sites where *Limax maximus* was recorded (i.e., present, black circle) or not recorded (absent, white circle), excluding the data on *L. maximus*: **a** results as determined from presence–absence data, **b** results as determined from quantitative data. Red and blue lines indicate the peripheries in the plots of sites with and without *L. maximus*, respectively

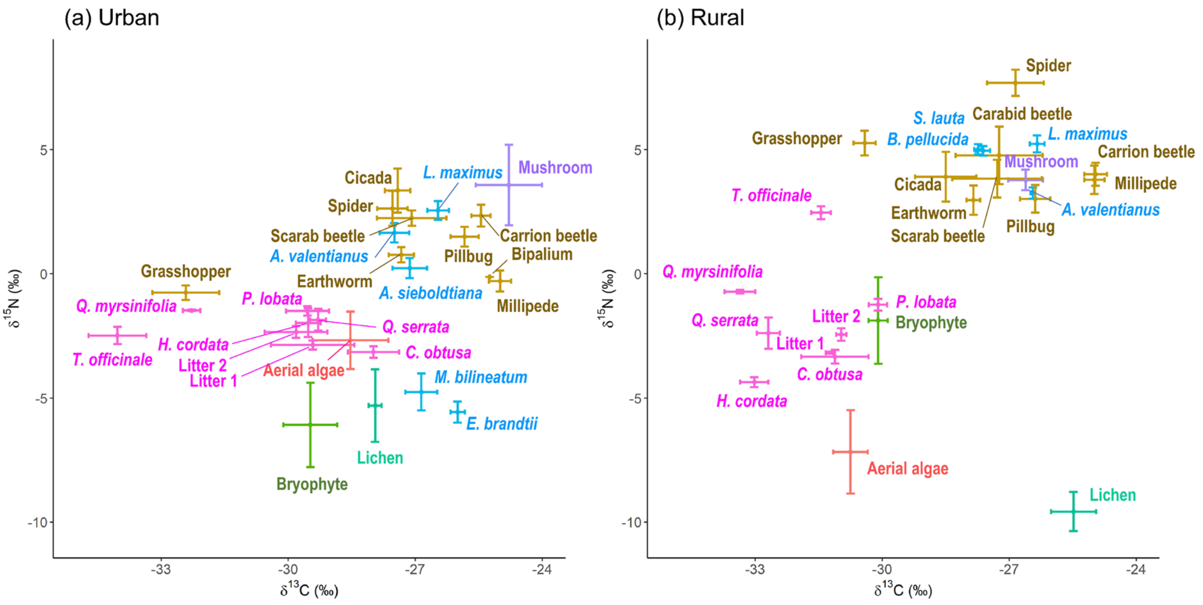


Fig. 5 Comparison of the stable carbon and nitrogen isotope ratios between **a** urban and **b** rural sites where *Limax maximus* was introduced. Colors indicate taxonomic differentiation: light blue: terrestrial gastropods; brown: invertebrates other than snails and slugs; pink: vascular plants; dark green: bryophytes; red: aerial algae; light green: lichens; purple, mushroom. See Supplementary Material 2 for the abbreviations of taxa

urban site was estimated to feed on animals, lichens, mushrooms and aerial algae in similar proportions (Fig. 6d).

Discussion

To identify the habitat and diet characteristics of *L. maximus* at sites where it has been introduced, we

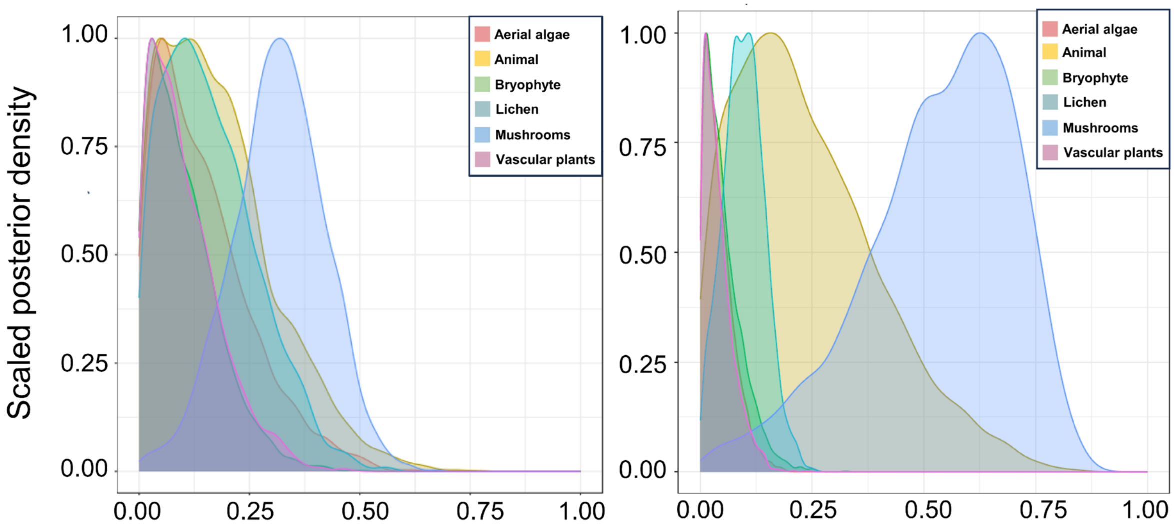
Table 1 Posterior distribution of diets of *Limax maximus*, *Ambigolimax valentianus*, and *Meghimatium bilineatum*, as estimated by MixSIAR on the basis of stable isotope analyses. Samples were collected in Ibaraki Prefecture, Japan, where *L. maximus* was introduced

	Aerial algae			Animals			Bryophyte			Lichen			Mushroom			Vascular plants		
	Min	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min	Mean	Max
Urban area																		
Limax maximus	0.004	0.121	0.418	0.006	0.156	0.475	0.003	0.083	0.293	0.009	0.145	0.394	0.103	0.317	0.508	0.004	0.085	0.306
Ambigolimax valentianus	0.007	0.149	0.455	0.011	0.181	0.449	0.007	0.117	0.354	0.01	0.17	0.435	0.012	0.149	0.341	0.007	0.146	0.448
Meghimatium bilineatum	0.003	0.087	0.419	0.002	0.056	0.255	0.006	0.169	0.625	0.034	0.451	0.79	0.004	0.078	0.285	0.002	0.05	0.224
Rural area																		
Limax maximus	0.001	0.033	0.125	0.013	0.21	0.61	0.002	0.04	0.156	0.013	0.097	0.191	0.141	0.557	0.787	0.002	0.032	0.124

applied two research approaches: field surveys and stable isotope analyses. By doing so, we were able to obtain several insights regarding the ecological impacts of invasion by *L. maximus*. Our field survey results indicated that *L. maximus* is a generalist that occupies a wide range of habitats from urban to rural environments (Fig. 2). The species is known as an agricultural pest (Kozłowski 2012; Stange and Deisler 2005), and it feeds on lettuce in vegetable fields in rural Japan (Iijima et al. 2013). Moreover, *L. maximus* prefers forest edges but does not necessarily need intact pristine forests as its habitat (Barker 1999). Likewise, we also frequently recorded the species at disturbed sites in urban and agricultural areas. In contrast, *A. valentianus*, another non-native slug that was introduced to Japan before *L. maximus*, occurred more dominantly in urban than in rural areas (Fig. 2), as described by Azuma (1982) and Namba et al. (2020). Because *L. maximus* has a much wider habitat range than this common non-native slug, it may share habitats with much higher numbers of native species, especially in rural areas.

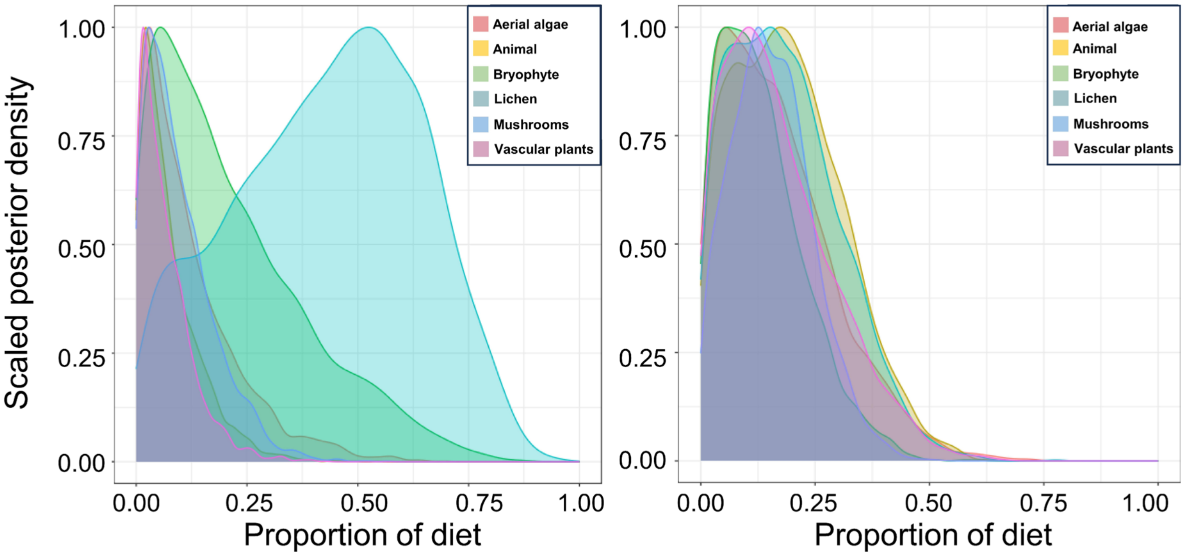
Although *Limax maximus* exhibits broad habitat occupancy and a flexible trophic niche, our analyses detected no significant differences in terrestrial molluscan community composition or diversity between sites where the species was present and those where it was absent. This indicates that, at the spatial scale examined, the establishment of *L. maximus* has not resulted in measurable assemblage-level restructuring. Traits such as habitat generalism, large body size, and omnivory may facilitate coexistence with resident species rather than direct displacement. Potential interactions at finer spatial or temporal scales cannot be excluded, but the absence of detectable community-level effects represents an ecologically meaningful outcome. These results highlight that widespread non-native species do not necessarily exert strong impacts on native assemblages and underscore the importance of documenting invasion scenarios characterized by low or context-dependent effects.

Stable isotope analyses indicated that the $\delta^{13}C$ and $\delta^{15}N$ values of *L. maximus* were similar to those of spiders, ground beetles, and carrion beetles (Fig. 5). Our finding agrees with those of a previous report on the stable isotope ratios of *L. maximus* in Hawaii (Meyer and Yeung 2011). The $\delta^{15}N$ of *L. maximus* in the urban area was significantly higher than those of



(a) *Limax maximus* (Urban)

(b) *Limax maximus* (Rural)



(c) *Meghimatium bilineatum* (Urban)

(d) *Ambigolimax valentianus* (Urban)

Fig. 6 Posterior density distributions comparing the variation in diet estimated by MixSIAR on the basis of stable isotope analyses. The x-axis indicates the proportion of diets, and the y-axis indicates the probability distribution estimated by running the Markov chain Monte Carlo simulation in MixSIAR based on the data collected by the isotope analyses. *Limax maximus* collected in **a** an urban area and **b** a rural area, **c** *Meghimatium bilineatum* collected in an urban area, **d** *Ambigolimax valentianus* collected in an urban area

the other terrestrial gastropods. Thus, *L. maximus* has a different trophic position from the native terrestrial gastropods in Japan. At the rural site, *L. maximus* had δ^{13}C and δ^{15}N values similar to those of *Succinea lauta* A. A. Gould, 1859 and *B. pellucida* which are known as herbivores by a previous study (Ikezawa 2012). Thus, the δ^{15}N values of these individuals were unexpectedly high. We are unable to identify the reasons for this seeming discrepancy, and some of the results from the rural site should be interpreted with caution. Nevertheless, a relatively high trophic position of *L. maximus* was observed consistently at the urban and rural sites.

MixSIAR analyses demonstrated that *L. maximus* is omnivorous, with a diet composed of principally mushrooms, along with animals, lichens, and other plant materials (Fig. 6). This estimate agrees with the trophic relationships described above (Fig. 5). Laboratory feeding experiments suggest that *L. maximus* feeds on plants (Joe 2006; Joe and Daehler 2008; Komatsu and Saeki 2022), but the proportion of plants estimated by the MixSIAR analyses was low. Thus, wild populations may be more dependent on food resources other than plants, or it is possible that C and N derived from non-plant resources, such as microbes, were assimilated (Meyer and Yeung 2011). In addition, the food dependency of *L. maximus* was more variable at the urban than at the rural site (Fig. 6). This implies that this species can change its diet flexibly in response to the availability of food resources, which is likely a factor in its rapid invasion and its rapid population growth at sites where it has been introduced. Unlike *L. maximus*, the other slug species were likely to be more dependent on lichens, bryophytes, and aerial algae, as previously reported from feeding experiments in the laboratory (Komatsu and Saeki 2022). In summary, *L. maximus* has a different diet from those of the other common slugs in Japan.

Management implications

Our analyses clearly demonstrated that *L. maximus* is a generalist with regard to habitat preference. We found that *L. maximus* shares a wide range of habitats with other terrestrial gastropods, and this characteristic is distinctively different from that of native slugs and a non-native slug previously introduced to Japan. In addition, *L. maximus* has a flexible diet: it feeds not only on plants but also on mushrooms and animals. Its trophic position was very different from that of the other terrestrial gastropods examined here. These characteristics probably contributed to its rapid population increase after its introduction to Japan, and its population will likely continue to expand in the future in both urban and rural ecosystems. Agriculture—including mushroom production—and horticulture are important in Japan, and effective measures to avoid the slug's population expansion are therefore urgently needed. To minimize agricultural and economic damage, we recommend that governmental, scientific, and other related organizations inform farmers about the ecological characteristics of *L. maximus*, such as its broad habitat range and preference for mushrooms, with the possibility of shifting its dietary habits. We also encourage these organizations to share the slug's occurrence information with the public. If citizens find this species at a very early stage of invasion, they can alert the government or scientific organizations immediately, at a relatively low cost (Hester and Chaco 2017; Morii et al. 2018). Then, an eradication program can be initiated soon after the introduction, when the population is still relatively small. Such a collaborative network could be an effective tool for avoiding or delaying the further range expansion of this invasive species.

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Data availability See Supplementary Material for the data collected by our field survey and stable isotope analyses.

Declarations

Conflict of interests The authors have no conflict of interest to disclose.

References

Aubry S, Labaune C, Magnin F, Kiss L (2005) Habitat and integration within indigenous communities of *Xeropicta derbentina* (Gastropoda: Hygromiidae) a recently introduced land snail in south-eastern France. Divers Distrib 11:539–547. https://doi.org/10.1111/j.1366-9516.2005.00189.x

Azuma M (1982) The land snails of Japan. Hoikusha, Osaka

Barker GM (1999) Naturalised terrestrial Stylommatophora (Mollusca: Gastropoda). Fauna N Z. https://doi.org/10.7931/J2/FNZ.38

Barker GM (2015) *Limax maximus* (leopard slug). CABI Compend. https://doi.org/10.1079/cabicompendium.3082

Barker GM, McGhie RA (1984) The biology of introduced slugs (Pulmonata) in New Zealand 1. Introduction and notes on *Limax maximus*. N Z Entomol 8:106–111. https://doi.org/10.1080/00779962.1984.9722482

Coote T, Loève E (2003) From 61 species to five: endemic tree snails of the Society Islands fall prey to an ill-judged biological control programme. Oryx 37:91–96. https://doi.org/10.1017/S0030605303000176

Cowie RH, Robinson AC (2003) The decline of native Pacific island faunas: changes in status of the land snails of Samoa through the 20th century. Biol Conserv 110:55–65. https://doi.org/10.1016/S0006-3207(02)00176-3

Gelman A, Rubin DB (1992) Inference from iterative simulation using multiple sequences. Stat Sci 7:457–511. https://doi.org/10.1214/ss/1177011136

Gerlach J, Barker GM, Bick CS et al (2021) Negative impacts of invasive predators used as biological control agents against the pest snail *Lissachatina fulica*: the snail *Euglandina* 'rosea' and the flatworm *Platydemus manokwari*. Biol Invasions 23:997–1031. https://doi.org/10.1007/s10530-020-02436-w

Hahn PG, Draney ML, Dornbush ME (2011) Exotic slugs pose a previously unrecognized threat to the herbaceous layer in a Midwestern woodland. Restor Ecol 19:786–794. https://doi.org/10.1111/j.1526-100X.2010.00710.x

Hasegawa K, Fukuda H, Ishikawa H (2009) Discovery of naturalized populations of the introduced slug, *Limax maximus* in Tsuchiura City, Ibaraki Prefecture, Japan. Newslett Malacol Soc Japan 39:101–105

Hester SM, Cacho OJ (2017) The contribution of passive surveillance to invasive species management. Biol Invasions 19:737–748. https://doi.org/10.1007/s10530-016-1362-4

Horsák M, Lososová Z, Čejka T et al (2013) Diversity and biotic homogenization of urban land-snail faunas in relation to habitat types and macroclimate in 32 Central European cities. PLoS ONE 8:e71783. https://doi.org/10.1371/journal.pone.0071783

Iijima K, Fukumoto M, Minato H (2013) Occurrence of the introduced slug, *Limax maximus* (Pulmonata: Limacidae) in Saku district, Nagano Prefecture, Japan. Kainakama 47:1–4

Ikezawa H (2012) Expansion of geographic distribution of *Bradybaena pellucida* in Ibaraki Prefecture and agricultural damage. A Museum 73:5

Joe S (2006) Impacts of alien slugs on native plant seedlings in diverse mesic forest, Oahu, Hawaii, and a study of slug food plant preferences. University of Hawaii, Manoa, Hawaii

Joe SM, Daehler CC (2008) Invasive slugs as under-appreciated obstacles to rare plant restoration: evidence from the Hawaiian Islands. Biol Invasions 10:245–255. https://doi.org/10.1007/s10530-007-9126-9

Keller HW, Snell KL (2002) Feeding activities of slugs on myxomycetes and macrofungi. Mycologia 94:757–760. https://doi.org/10.2307/3761690

Kimura K, Chiba S (2010) Interspecific interference competition alters habitat use patterns in two species of land snails. Evol Ecol 24:815–825. https://doi.org/10.1007/s10682-009-9339-8

Komatsu W, Saeki I (2022) Feeding habits of the exotic invasive slug *Limax maximus*: a basis for risk assessment of herbivory on forest and agricultural ecosystems. Manag Biol Invasions 13:644–658. https://doi.org/10.3391/mbi.2022.13.4.04

Kozłowski J (2012) The significance of alien and invasive slug species for plant communities in agrocenoses. J Plant Prot Res 52:67–76. https://doi.org/10.2478/v10045-012-0012-9

Krebs CJ (1999) Ecological Methodology. Addison Wesley Longman, Menlo Park

Kurozumi T (2002) *Ambigolimax valentianus* In: Ecological Society of Japan (ed) Handbook of alien species in Japan. Chijinshokan, Tokyo, pp 164

McCue MD, Javal M, Clusella-Trullas S et al (2020) Using stable isotope analysis to answer fundamental questions in invasion ecology: progress and prospects. Methods Ecol Evol 11:196–214. https://doi.org/10.1111/2041-210X.13327

Meyer WM, Yeung NW (2011) Trophic relationships among terrestrial molluscs in a Hawaiian rain forest: analysis of carbon and nitrogen isotopes. J Trop Ecol 27:441–445. https://doi.org/10.1017/S0266467411000058

Mito T, Uesugi T (2004) Invasive alien species in Japan : the status quo and the new regulation for prevention of their adverse effects. Glob Environ Res 8:171–193

Morii Y, Yokoyama J, Kawata M et al (2015) Evidence of introgressive hybridization between the morphologically divergent land snails *Ainohelix* and *Ezohelix*. Biol J Linn Soc 115:77–95. https://doi.org/10.1111/bij.12466

Morii Y, Okubo Y, Watanabe S (2018) Activity of invasive slug *Limax maximus* in relation to climate conditions based on citizen's observations and novel regularization based statistical approaches. Sci Total Environ 637:1061–1068. https://doi.org/10.1016/j.scitotenv.2018.04.403

Namba N, Mukaimine W, Haga T, Saeki I (2020) Terrestrial and freshwater mollusks recorded from the University of Tsukuba Campus, Tsukuba City, Japan. Newslett Malacol Soc Japan 50:221–240

Němec T, Líznarová E, Birkhofer K, Horsák M (2021) Stable isotope analysis suggests low trophic niche partitioning among co-occurring land snail species in a floodplain forest. J Zool 313:297–306. https://doi.org/10.1111/jzo.12859

Norden A, Williams G (2015) *Limax maximus* (Gastropoda: Limacidae) predation on *Magicicada septendecim* (Hemiptera: Cicadidae) in Prince Georges County, Maryland. Md Nat 53:52–54

O'Hanlon A, Williams CD, Gormally MJ Terrestrial slugs (Mollusca: Gastropoda) share common anti-predator defence mechanisms but their expression diff ers among species. J Zool 307:203–214. https://doi.org/10.1111/jzo.12635

Oksanen J, Simpson G, Blanchet F, Kindt R, Legendre P, Minchin P, O'Hara R, Solymos P, Stevens M, Szoecs E, Wagner H, Barbour M, Bedward M, Bolker B, Borcard D, Carvalho G, Chirico M, De Caceres M, Durand S, Evangelista H, FitzJohn R, Friendly M, Furneaux B, Hannigan G, Hill M, Lahti L, McGlinn D, Ouellette M, Ribeiro Cunha E, Smith T, Stier A, Ter Braak C, Weedon J (2022) Vegan: community Ecology Package. https://CRAN.R-project.org/package=vegan

Paustian ME, Barbosa P (2011) Overlap of food and microhabitat preferences among some native and nonnative slugs in mid-Atlantic forests of eastern North America. J Molluscan Stud 78:92–99. https://doi.org/10.1093/mollus/eyr041

Post DM (2002) Using stable isotopes to estimate trophic position: models, methods, and assumptions. Ecology 83:703–718. https://doi.org/10.1890/0012-9658(2002)083[0703:USITET]2.0.CO;2

R Core Team (2022) A language and environment for statistical computing. In: R foundation for statistical computing. https://www.R-project.org/

Rollo CD, Wellington WG (1979) Intra- and inter-specific agonistic behavior among terrestrial slugs (Pulmonata: Stylommatophora). Can J Zool 57:846–855. https://doi.org/10.1139/z79-104

Roth B, Sadeghian PS (2006) Checklist of the land snails and slugs of California. Second Edition. Santa Barbara Museum of Natural History, Santa Barbara, California

Sessions L, Kelly D (2002) Predator-mediated apparent competition between an introduced grass, *Agrostis capillaris*, and a native fern, *Botrychium australe* (Ophioglossaceae), in New Zealand. Oikos 96:102–109. https://doi.org/10.1034/j.1600-0706.2002.960111.x

Stange LA, Deisler JE (2005) Slugs of Florida (Gastropoda: Pulmonata): EENY-087/IN244, rev. 4/2004. EDIS 2005

Stock BC, Jackson AL, Ward EJ et al (2018) Analyzing mixing systems using a new generation of Bayesian tracer mixing models. PeerJ 6:e5096. https://doi.org/10.7717/peerj.5096

Takashima H (1954) Considerations on the change of animal life in Japan. J Yamashina Inst Ornithol 1:146–155

Takeuchi K (2001) Nature conservation strategies for the 'SATOYAMA' and 'SATOCHI', habitats for secondary nature in Japan. Glob Environ Res 5:193–198

Taylor J (1902) Monograph of the land and freshwater Mollusca of the British Isles part 8. Leeds, Taylor Brothers

Tomiyama K (2002) Land Snails of the Ogasawara Islands: Fragile Oceanic Island Endemic Species and the Causes of Their Extinction. Forest Sci (Shinrin Kagaku) 34:25–28 (in Japanese)

Udaka T, Tanaka H (2010) Slugs. Nousangyosonbunkakyoukai. Japan

Watz J, Johansson S, Nyqvist D (2022) Egg production in *Arion vulgaris*: density dependence in A. vulgaris and interspecific effects of *Limax maximus*. J Molluscan Stud 88(3):eyac026. https://doi.org/10.1093/mollus/eyac026