



RESEARCH ARTICLE

# Effect of Asian clam shells on aquatic fauna in an artificial ditch

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## Abstract

Freshwater bivalves perform a range of ecological functions in both rivers and lakes. However, the effects of these functions in artificial ditches, one of the main habitats for bivalves, have not yet been examined. Among the burrowing bivalves, the shells of the Asian clam, genus *Corbicula*, decay slowly underwater and often dominate the bottom of natural rivers and artificial ditches. Here, a manipulative experiment was performed in a mesocosm ditch to determine the differences in the abundance and community composition of aquatic fauna in cages with and without *Corbicula* shells and to identify the relationship between shell size composition and fauna. Following the experiment, kick net sampling was performed to identify the fauna in the ditch. The results of the experiment indicated that macroinvertebrate species richness was higher in cages with a mix of small and large shells than in those without shells. The total number of macroinvertebrates was higher in cages with shells than in those without shells. However, both the species richness and total number of macroinvertebrates did not significantly differ among the treatments with different shell size compositions. Kick net sampling revealed macroinvertebrate fauna and the presence of a fish species in the mesocosm. These results suggested that *Corbicula* shells may be useful for the conservation of macroinvertebrates in artificial ditches with concrete-lined and simple structures.

**Keywords** Anthropogenic habitat · Conservation · *Corbicula* · Ecosystem engineering · Freshwater bivalve · Reconciliation ecology

## Introduction

Freshwater-burrowing bivalves, including the genus *Corbicula* and order Unionoida, have important ecological functions in rivers and lakes (Sousa et al. 2009; Vaughn 2018; Vaughn and Hoellein 2018). The physical presence of living bivalves and their spent shells results in habitat complexity and heterogeneity (Vaughn and Hakenkamp 2001; Gutiérrez et al. 2003; Strayer et al. 2004; Bódis et al. 2014; Ilarri et al. 2018). Bivalve biofiltration can remove substantial quantities of particles from the water column and alter the water quality (Welker and Walz 1998; Vaughn et al. 2008; Pigneur et al. 2014; Choudhury et al. 2016). Bivalve excretion influences ecosystem processes by modifying nutrient cycling (Atkinson et al. 2013) as bivalves excrete a considerable amount of feces and pseudofeces (Strayer 2014; Vaughn 2018), and this deposition is partially responsible for the

growths of benthic algae, macroinvertebrates, and lampreys (Howard and Cuffey 2006; Spooner and Vaughn 2006; Limm and Power 2011; Allen et al. 2012; Strayer 2014; Atkinson et al. 2021; Simeone et al. 2021). Bioturbation occurs when bivalves move and feed, altering the sediment water content, oxygen penetration, solute flux, and sediment nitrate, chloride, and calcium carbonate concentrations (Vaughn and Hakenkamp 2001).

Bivalves generally produce large amounts of shells, either individually or when aggregated in dense beds. These shells alter the benthic environments and are thus, important elements in a habitat structure, as they support the maintenance of high species richness, particularly in soft-bottomed environments (Gutiérrez et al. 2003; Sousa et al. 2009; Ilarri et al. 2019). Shells are substrata for the attachment of epibionts, thereby providing refuges from predation and disturbance and controlling the transport of resources in the benthic environment (Gutiérrez et al. 2003). They persist after the bivalve has died and retain their ecological function (Gutiérrez et al. 2003; Strayer and Malcom 2007; Ilarri et al. 2015b, 2019).

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Bivalves inhabit and can often be found in abundance in artificial habitats, such as ditches, including canals and navigations, predominantly for agricultural use (Araujo and Ramos 2000; Gómez and Araujo 2008; Negishi et al. 2011; Sousa et al. 2019, 2021). However, their ecological functions in ditches have yet to be explored. Ditches frequently harbor uncommon biota and endangered species (Herzon and Helenius 2008; Chester and Robson 2013; Katayama et al. 2015). Ditches often have high beta diversity of aquatic organisms, such as fish and invertebrates, which in turn contribute to gamma diversity at a landscape scale (Armitage et al. 2003; Williams et al. 2004; Verdonschot et al. 2011; Hill et al. 2016; Ishiyama et al. 2016). However, many traditional ditches with natural bottom and side structures are concrete-lined with simple structures, which are less suitable habitats for aquatic organisms (Katano et al. 2003; Gómez and Araujo 2008; Katayama et al. 2015; Sousa et al. 2021).

The Asian clam genus *Corbicula* originated in Asia, Australia, and Africa (Sousa et al. 2008). *Corbicula* has rapidly expanded its distribution worldwide (Crespo et al. 2015) and is considered an opportunistic species because of its short life cycle, rapid growth, early sexual maturity, and high fecundity (Sousa et al. 2008, 2014). The shell of *Corbicula* is thicker and harder than that of other freshwater bivalves, such as unionids. Therefore, it decays more slowly underwater (Strayer and Malcom 2007; Schmidlin et al. 2012; Ilarri et al. 2015b, 2019). *Corbicula* shells often dominate the shells that have accumulated in the beds of rivers and lakes, thereby providing a higher level of ecological function than other species (Strayer and Malcom 2007; Ilarri et al. 2019). Overall, *Corbicula* is widely distributed in artificial ditches (Sousa et al. 2021). Given the important role of shells in natural rivers and lakes, the presence of *Corbicula* shells may strengthen habitat complexity and heterogeneity in artificial ditches, thereby supporting aquatic fauna. However, to the best of my knowledge, this has not yet been demonstrated.

This study aimed to explore whether microhabitat complexity and heterogeneity created by *Corbicula* shells positively affect aquatic fauna in artificial ditches. An experiment was performed in a mesocosm ditch mimicking agricultural ditches to (i) determine the difference in aquatic fauna between microhabitat patches (cages) with and without shells and (ii) identify the relationship between shell size composition and fauna because the ecological effects may differ depending on shell size composition (Ilarri et al. 2015a, 2018).

## Methods

### Study species

*Corbicula* was selected because it is one of the most representative bivalve species in artificial ditches (Nakano and

Morii 2019; Sousa et al. 2021). *Corbicula fluminea* (Müller 1774) is considered non-native to Japan and is currently distributed throughout the Japanese archipelago (Okawa et al. 2016). However, *Corbicula fluminea* and native *C. leana* Prime, 1864 cannot be distinguished based on their morphology and genetics. It is likely that *C. leana* is a population of *C. fluminea* (Komaru et al. 2012, 2013; Wang et al. 2014), and so all shells in the present study were allocated to the genus *Corbicula*.

## Experiment

Intact shells (with both sides attached) of dead *Corbicula* were collected on April 25, 2021, from an artificial ditch (31°43' N, 131°04' E) in Miyakonojo City, Miyazaki Prefecture, southern Kyushu Island, Japan. The ditch had a live *Corbicula* density of 5350 individuals·m<sup>-2</sup> and an empty shell density of 3825 individuals·m<sup>-2</sup> on the sampling day. In the laboratory of Minami Kyushu University, Miyazaki Prefecture, shells were cleaned using brushes to remove any attached matter including the soft tissues of *Corbicula*. The samples were then dried at 50 °C for 63 h and weighed to the nearest 0.1 g. Their shell lengths were measured to the nearest 0.05 mm using a caliper.

The surface area of each shell was estimated in accordance with previous studies (Beckett et al. 1996; Werner and Rothhaupt 2007; Bódis et al. 2014). Thirty shells (shell length range 13.15–40.10 mm) collected from the ditch were wrapped and fitted with aluminum foil to estimate the shell surface area, with the outside and inside of each shell being fitted separately. The foil masses on both sides were measured to a precision of 0.0001 g using an electronic analytical balance (METTLER TOLEDO Inc., type: AG204). Subsequently, a foil mass-to-area regression curve was constructed ( $y = 2.1941x - 23.782$ ,  $R^2 = 0.9817$ ).

The manipulative experiment was performed in a mesocosm, which was an artificial ditch that was constructed in 2009 for study and education in an outside field at Minami Kyushu University (Fig. 1). The mesocosm was 30 m long, 0.8–4.0 m wide of water surface, and with up to 20 cm of water depth, and a flow velocity reaching up to 0.6 cm·s<sup>-1</sup>. The base and sides of the mesocosm were lined with concrete, and sediment accumulated at the concrete base. The mesocosm was divided into five parts with four small waterfalls (8–30 cm in height) that mimicked the waterfalls in agricultural ditches (Fujimoto et al. 2008). The underground water supplied by a pump flowed continuously throughout the year. There were no bivalves inhabiting the ditch, and the only aquatic plant observed was the crystalwort *Riccia*. Insects, including dragonflies (order Odonata), and frog *Glandirana rugosa* (Temminck et Schlegel 1838) spawn annually in the mesocosm. Although there was no record





**Fig. 1** Photographs of the manipulative experiment. The mesocosm ditch considered for experimentation (**a** taken from upstream on September 30, 2022). The downstream section where the experiment was

performed (**b** taken from downstream on September 30, 2022, **c** taken on May 23, 2021 during the experiment period). Stainless cage used in the experiment (**d** from the side, **e** from the top)

of release history in the ditch before 2019, other aquatic organisms, such as gastropods, crustaceans, and fish, were believed to have been introduced into the mesocosm until 2019 after which they colonized (see results for detailed biota). Because the mesocosm was not connected to any other waterbodies, they could not invade on their own. Moreover, no organisms were released after 2020.

The experiment was performed between May 23 and June 20, 2021. This period was selected because aquatic animals, including dragonfly larvae and tadpoles, were abundant in

previous years (Nakano, unpublished data). The water temperatures at the start and end of the experiment were 25.0 and 22.7 °C, respectively. A total of 16 stainless cages, each with 22.5 cm diameter, 11.2 cm height, 0.2 cm mesh size, and an open top were placed in the mud at the bottom of the downstream section (8.6 m long). The surface area of the mud in each cage was 0.0254 m<sup>2</sup> and the cages were located approximately 50 cm apart. They were filled with mud to a depth of 5 cm and buried 5 cm into the mesocosm substrate so that the sediment in the cage was level with that of the

mesocosm. This left the approximately upper 6 cm of the cage extending into the water column. This design allowed aquatic organisms to access the cages from the open tops but prevented the immigration/emigration of *Corbicula* shells to maintain constant shell densities over time.

The experimental design comprised four shell treatments, each with a constant shell weight across treatments, namely, large shells, small shells, and a mix of large and small shells, along with a shell-free control with only sediment, comprising four replications for each. Sixteen cages were used in the study, and each cage was assigned to one of the treatment groups. The shell lengths for the large and small shells were 28.3–40.4 and 13.2–20.6 mm, respectively. The number and surface area of shells per cage were determined using a constant dry weight for each cage (mean  $\pm$  standard deviation:  $11.4 \pm 0.8$  g;  $450.1 \pm 32.8$  g  $\cdot$  m<sup>-2</sup>) (Table 1). The mean  $\pm$  standard deviation of the area per cage were  $193.8 \pm 6.0$  cm<sup>2</sup> ( $7631.3 \pm 235.8$  cm<sup>2</sup>  $\cdot$  m<sup>-2</sup>) in the large,  $253.5 \pm 3.9$  cm<sup>2</sup> ( $9979.8 \pm 153.8$  cm<sup>2</sup>  $\cdot$  m<sup>-2</sup>) in the small, and  $220.0 \pm 14.1$  cm<sup>2</sup> ( $8662.5 \pm 554.8$  cm<sup>2</sup>  $\cdot$  m<sup>-2</sup>) in the mixed treatments. All cages were placed in the mesocosm ditch for 28 d (May 23–June 20). At the end of the experiment, the cage contents were collected and sieved using a 0.5 mm mesh sieve. The organisms were sorted, fixed in 99% ethanol, identified to the lowest practical taxonomic level using a binocular stereomicroscope, and counted in the laboratory.

To identify fauna in the mesocosm during the experimental period, samples were collected immediately after the end of the experiment on June 20. The aquatic organisms were collected by two people using kick nets with a 1 mm mesh size for 15 min in the sediment and water

column in the downstream section of the mesocosm. Subsequently, the organisms were sorted, fixed in 99% ethanol, identified to the lowest practical taxonomic level using a binocular stereomicroscope, and counted in the laboratory.

## Data analysis

To estimate the effect of shell treatments on aquatic fauna, generalized linear models (GLMs) were fitted to the data using Poisson distribution. A negative binomial distribution was not used because the *R*-squared values of the models using this distribution were lower than those of the models using Poisson distribution. Each model included one of the objective variables, namely species richness or the total number of individuals for all species, and the shell treatment as the categorical explanatory variable with four categories of large, small, mixed, and control treatments. The *R*-squared value for each GLM was calculated to determine whether shell presence was the main factor affecting the aquatic fauna in the cages. The number of individuals per species was not used as an objective variable because it was too low to indicate significant results, regardless of the species.

All analyses were performed using R software (version 4.0.0; R Core Team 2020). The glm function of the stats package was used to construct the GLMs, the r2 function of the performance package was used for calculating *R*<sup>2</sup>, and the confint function of the stats package was used to estimate the 95% confidence limits.

**Table 1** Treatments for the manipulative experiment in the mesocosm ditch

	Homogenous Large	Small	Heterogeneous Mix
Shell length (mm)			
Range	28.3–37.7	13.2–20.6	14.7–40.4
Mean	32.9	16.6	19.2
SD	2.7	1.8	6.4
Density of shells per cage N $\cdot$ m <sup>-2</sup>	157	787	472
Shell surface area per cage			
Range (cm <sup>2</sup> $\cdot$ m <sup>-2</sup> )	6957.5–8080.5	9322.2–10535.9	8079.4–9452.9
Mean (cm <sup>2</sup> $\cdot$ m <sup>-2</sup> )	7631.3	9979.8	8662.5
SD (cm <sup>2</sup> $\cdot$ m <sup>-2</sup> )	235.8	153.8	554.8
Dry weight per cage			
Range (g $\cdot$ m <sup>-2</sup> )	429.1–472.4	417.3–492.1	405.5–507.9
Mean (g $\cdot$ m <sup>-2</sup> )	449.8	453.7	446.9

The *Corbicula* shells for each treatment with large shells, small shells, mix of large and small shells are shown. Each treatment had four replications. Shells were put on mud across a surface area of 0.0254 m<sup>2</sup> in each cage. The number and surface area of shells per cage were determined using a constant dry weight of shells

SD standard deviation

## Results

A total of 337 individuals from 10 taxa were observed in the cages (Table 2). Seven macroinvertebrate taxa, namely the tadpole *G. rugosa*, isopod *Asellus hilgendorfi* Bovallius, 1886, chironomid Chironomidae larvae, crane fly Tipulidae larvae, dragonfly *Sympetrum eroticum* (Selys 1883) larvae, dragonfly Coenagrionidae larvae, and worm Oligochaeta, were only present in cages that contained *Corbicula* shells. Three taxa, namely the gastropod *Semisulcospira libertina* (Gould 1859), shrimp *Neocaridina*, and dragonfly Libellulidae larvae, were present in cages with and without *Corbicula* shells.

The model results indicated that the species richness and total number of macroinvertebrates differed significantly among the treatments. Specifically, the species richness was significantly higher in cages with mixed-sized shells than in those without shells (Table 3). The effects of homogenous large and small shell size treatments on species richness were not significant. The ranges of species richness partly overlapped among the large, small, and mixed shell size treatments (Fig. 2), and there was no clear difference in species richness among these treatments.

The total number of macroinvertebrates was significantly higher in cages with shells than in those without shells. The effects of all the treatments with shells on the total number of macroinvertebrates were significant. The ranges of the total number of macroinvertebrates widely overlapped among these treatments; therefore, there was no clear difference in total number of macroinvertebrates among the treatments with shells. Kick net sampling in the mesocosm resulted in the capture of 177 individuals from eight taxa (Table 2). One fish species, the Chinese minnow *Rhynchocypris oxycephalus jouyi* (Jordan et Snyder 1901), which was not present in any cage in the experiment, was captured using this method. The gastropod *S. libertina* was the most abundant species, accounting for 55% of the total (98 gastropods/177 all species), followed by the shrimp *Neocaridina*, which formed 26% of the total (46 shrimps/177 all species). These results along with the experimental results indicated that Chironomidae, Tipulidae, and Oligochaeta were present in cages with *Corbicula* shells. However, these organisms were not present in cages without *Corbicula* shells, and were not collected by kick net sampling in the mesocosm.

**Table 2** Model results from the manipulative experiment in the mesocosm ditch

Taxon	Large		Small		Mix		Control		Kick net
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	
Species richness	3.3	1.5	4.0	1.4	5.3	1.3	2.0	0.8	8
All species	22.5	13.3	27.3	11.1	23.5	12.4	11.0	8.0	177
Mollusca									
<i>Semisulcospira libertina</i>	18.5	11.6	21.3	8.5	16.3	10.1	9.5	7.0	98
Amphibia									
<i>Glandirana rugosa</i> <sup>a</sup>	0.3	0.5	0.0	0.0	0.0	0.0	0.0	0.0	17
Crustacea									
<i>Asellus hilgendorfi</i>	1.0	0.8	0.8	1.0	1.3	1.0	0.0	0.0	1
<i>Neocaridina</i>	0.5	1.0	1.8	1.7	0.5	0.6	1.3	1.0	46
Insecta									
Chironomidae <sup>a</sup>	2.0	1.8	2.8	1.3	2.5	1.7	0.0	0.0	0
Tipulidae <sup>a</sup>	0.0	0.0	0.0	0.0	1.8	1.3	0.0	0.0	0
Libellulidae <sup>a</sup>	0.3	0.5	0.5	0.6	0.3	0.5	0.3	0.5	8
<i>Sympetrum eroticum</i> <sup>a</sup>	0.0	0.0	0.3	0.5	0.3	0.5	0.0	0.0	2
Coenagrionidae <sup>a</sup>	0.0	0.0	0.0	0.0	0.5	0.6	0.0	0.0	4
Oligochaeta									
sp 1	0.0	0.0	0.0	0.0	0.3	0.5	0.0	0.0	0
Fish									
<i>Rhynchocypris oxycephalus jouyi</i>	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1

Means and standard deviations (SD) of the species richness and numbers of individuals of both all species and each species are shown. The experiment was performed between May 23 and June 20, 2021. The experimental design comprised four treatments, namely large, small, mixed shell size treatments, and a shell-free control treatment. Each treatment had four replications. Kick net collection was performed in the ditch immediately after the end of the experiment on June 20

<sup>a</sup>Larvae

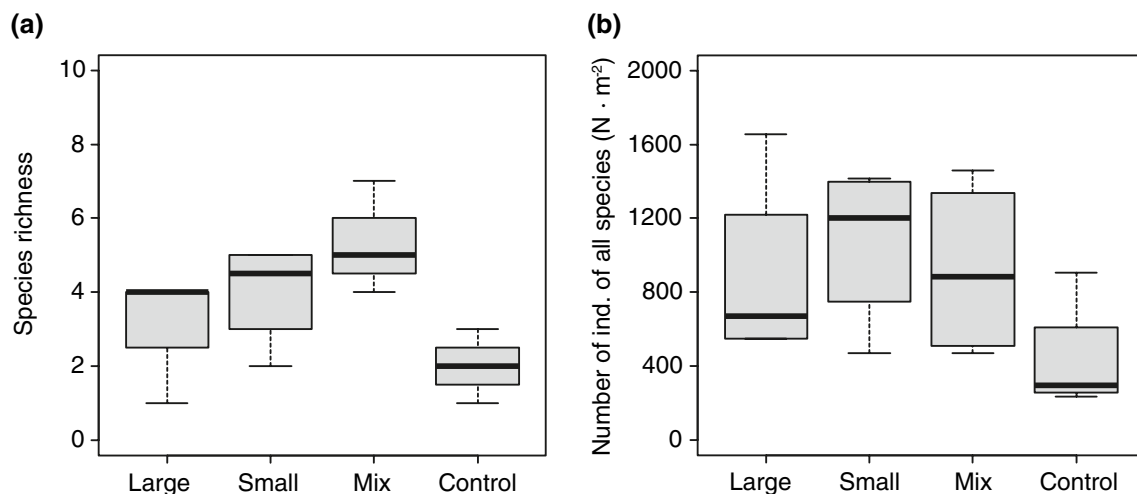


**Table 3** Model results from the manipulative experiment in the mesocosm ditch

Objective variable	Estimate	SE	<i>P</i>	95% CL		<i>R</i> -squared
				2.5%	97.5%	
Species richness						0.601
(Intercept)	0.69	0.35	0.050	-0.09	1.31	
[Large]	0.49	0.45	0.280	-0.38	1.41	
[Small]	0.69	0.43	0.109	-0.13	1.60	
[Mix]	0.97	0.42	0.020	0.19	1.84	
Total number of individuals of all species						0.861
(Intercept)	2.40	0.15	<0.001	2.09	2.68	
[Large]	0.72	0.18	<0.001	0.36	1.08	
[Small]	0.91	0.18	<0.001	0.56	1.27	
[Mix]	0.76	0.18	<0.001	0.41	1.13	

Generalized linear models using a Poisson distribution were constructed. Each model included one of the objective variables, namely species richness or the total number of individuals of all species, and the shell treatment as the categorical explanatory variable with the four categories of large, small, mixed, and the control treatments

*Estimate* estimate coefficient, *SE* standard error, *P* Wald test *P* value, *CL* confidence limits



**Fig. 2** Box plots of the results of the manipulative experiment in a mesocosm. The lower ends, top ends, central parts, and error bars represent 25 and 75% quartile ranges, medians, and ranges. *Large*

*large shells*, *Small*: small shells, *Mix* mix of large and small shells, *Control* no shells, *ind.* individuals

## Discussion

*Corbicula* is frequently abundant in artificial ditches, and mainly agricultural ditches. To determine whether the microhabitat complexity and heterogeneity created by *Corbicula* shells affect the aquatic fauna in artificial ditches, a manipulative experiment was performed in a mesocosm ditch with a narrow width, shallow water depth, concrete-lined simple bottom, and side structures in the same way as modern agricultural ditches (Negishi et al. 2011; Katayama et al. 2015; Sousa et al. 2019). The aquatic fauna in agricultural ditches was well reproduced in the mesocosm

because aquatic organisms, such as gastropods, tadpoles, shrimps, dragonfly larvae, chironomid larvae, and cyprinid minnows, inhabiting the mesocosm are widely distributed in agricultural ditches (Katano et al. 2003; Watson and Ormerod 2004; Herzon and Helenius 2008; Amano et al. 2010; Katayama et al. 2015). Therefore, the results of this study are potentially applicable to modern agricultural ditches and other artificial ditches under similar abiotic and biotic conditions.

The results of the manipulative experiment indicated that the species richness and total number of macroinvertebrates were significantly higher in cages with *Corbicula* shells than in those without the shells, with the exception of the species

richness in the homogenous large and small shell size treatments. Chironomidae larvae, Tipulidae larvae, and Oligochaeta were only present in cages with *Corbicula* shells. The species richness and total macroinvertebrates did not clearly differ among the large, small, and mixed shell size treatments, indicating that the effects of size composition, surface area, and number of shells on macroinvertebrates were not evidently detected. Results from the kick net collection revealed macroinvertebrate fauna in the mesocosm as well as the presence of the Chinese minnow *R. o. jouyi*. To the best of my knowledge, no previous study has explored the ecological function of shells in artificial ditches. The results of the present study were then compared with those of previous studies performed in natural rivers and lakes. This comparison was required to clarify the differences in shell function between natural and artificial habitats.

The species richness and total number of macroinvertebrate individuals were mostly more abundant in cages with shells than in those without shells. This finding was consistent with the results of previous studies performed in natural habitats. Werner and Rothhaupt (2007) showed that the density of mayfly larvae increased in boxes with empty shells of *C. fluminea* compared to boxes containing live *C. fluminea* or only lake sand. Bódis et al. (2014) found that empty bivalve shells positively affected the presence of amphipods, caddis larvae, and isopods in a river, and highlighted that the effects of non-native the pond mussel *Sinanodonta woodiana* (Lea 1834) and *C. fluminea* shells on the habitat were particularly important for macroinvertebrate communities. The present study suggested that the presence of *Corbicula* shells positively affects the species richness and number of macroinvertebrates in both natural habitats and artificial ditches.

Although effects of shells on macroinvertebrates were detected, the main mechanisms were not determined in the present study. Shells generally provide resources and refuges to avoid physical and physiological disturbances and predation (Gutiérrez et al. 2003). Thus, the possible mechanisms responsible for the results of the present study may include providing refuges for macroinvertebrates to avoid predation because a potential predator is the omnivorous fish *R. o. jouyi* which inhabits the mesocosm. However, this remains speculative, and future studies are required to confirm this possibility and other potential mechanisms.

The species richness did not clearly differ among the treatments with different shell size compositions. This result was consistent with that of Ilarri et al. (2018), indicating that the species richness of macroinvertebrates in a river did not vary among shell treatments containing homogenous and heterogeneous bivalve species. In contrast, Ilarri et al. (2015a) found that macroinvertebrate species richness was higher with large unionid shells than with small *C. fluminea* shells or with both unionid and *C. fluminea* shells in a river.

Therefore, the relationship between shell size composition and species richness is likely to differ among habitats with different biotic and abiotic conditions. Although Ilarri et al. (2015a) performed an interspecific comparison between *C. fluminea* and unionids, the present study performed an intraspecific comparison of *Corbicula* shells. This may be one of the reasons for the different results between the studies. Additional studies are needed to determine these, especially in artificial ditches, where studies on the ecological function of bivalves are scarce.

The total number of macroinvertebrates were more abundant in cages with shells than in those without shells. The dominant species was the gastropod *S. libertina*, which accounted for over 60% of all the species in each cage in all treatments. *Corbicula* shells are likely to provide an available surface for *S. libertina* attachment. Ilarri et al. (2018) suggested that the overall dominance of the faucet gastropod *Bithynia tentaculata* (Linnaeus 1758) was observed for all shell treatments. They highlighted that several physical and biological characteristics provided by the deposition of bivalve shells were positively related to the number of gastropods. This may also be applicable to *libertina* in artificial ditches.

Ilarri et al. (2018) showed that the substrate complexity created by bivalve shells affects the macroinvertebrate density in a river, with treatments containing heterogeneous shells from multiple species yielding significantly denser macroinvertebrate assemblages than homogeneous shells from the treatment with one species. However, the results of the present study differed from those of Ilarri et al. (2018). This is because there was no clear difference in macroinvertebrate density among the treatments with different shell size compositions. As was the case with the species richness results, this inconsistency between studies suggested that the ecological function of *Corbicula* shells may differ among habitats under different biotic and abiotic conditions and that the effect of the size composition of simple *Corbicula* species was weak (or nothing) compared with the effect of shell size composition of multiple species.

Artificial ditches suitable for aquatic organisms generally harbor uncommon biota and endangered species (Herzon and Helenius 2008; Chester and Robson 2013; Katayama et al. 2015). Ditches often have high beta diversity of aquatic organisms, such as fish and invertebrates, which in turn contribute to gamma diversity at a landscape scale (Armitage et al. 2003; Williams et al. 2004; Verdonschot et al. 2011; Hill et al. 2016; Ishiyama et al. 2016). However, many traditional ditches with natural bottom and side structures have been reconstructed into modern ditches with concrete-lined and simple structures, which are less suitable habitats for aquatic organisms (Katano et al. 2003; Gómez and Araujo 2008; Katayama et al. 2015; Sousa et al. 2021). As suggested in the present study, *Corbicula* shells should be used for the

conservation of macroinvertebrates in modern ditches. For example, cleaning and maintenance activities are undertaken in ditches without focusing on biodiversity, consequently, resulting in the removal of sediments and mussels from ditches (Sousa et al. 2019, 2021). When these activities are undertaken, the remaining *Corbicula* shells may be effective in conserving macroinvertebrates in ditches. However, *Corbicula* can rapidly expand its distribution (Crespo et al. 2015). The colonization of non-native *Corbicula* often reduces the population of native unionids, and causes socio-economic damages (Yeager et al. 1999; Sousa et al. 2008, 2009; Ferreira-Rodríguez et al. 2018; Haag et al. 2020). Therefore, the introduction of live *Corbicula* in uncolonized areas should be avoided.

Because the experiment in the present study was performed in one mesocosm, it is important to explore the effect of shells on the aquatic fauna in artificial ditches under different biotic and abiotic conditions from the mesocosm ditch. Water depth, flow velocity, sediment, and vegetation often differ among ditches (Katano et al. 2003; Ohira et al. 2015; Nakano et al. 2017). Although the fauna and physical conditions of agricultural ditches were well reproduced in the mesocosm ditch, paddy drainage water, pesticides, and fertilizers generally inflow into agricultural ditches (Katayama et al. 2015; Nakano 2017) and may affect shell function. However, this was not determined in the present study and should be explored in the future.

## Conclusion

*Corbicula* shells are widely distributed in artificial ditches. Given the important role of shells in natural rivers and lakes, the presence of *Corbicula* shells may strengthen habitat complexity and heterogeneity in artificial ditches, thereby supporting aquatic fauna. However, this possibility has not been examined as yet. To determine whether the microhabitat complexity and heterogeneity created by *Corbicula* shells affect aquatic fauna in ditches, a manipulative experiment was performed in a mesocosm ditch with physical conditions and fauna similar to agricultural ditches. The results of the experiment indicated that the species richness and total number of macroinvertebrates were mostly higher in cages with *Corbicula* shells than in those without the shells. This study provided evidence for the first time that *Corbicula* shells can be used for the conservation of macroinvertebrates in ditches. For example, when cleaning and maintenance activities are undertaken in ditches, the remaining *Corbicula* shells could be effective in conserving macroinvertebrates. The ecological effect of *Corbicula* shells may be particularly important in modern ditches with concrete-lined and simple structures, which are unsuitable habitats for aquatic organisms.

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**Data availability** The datasets generated and/or analyzed during the present study are available from the corresponding author upon reasonable request.

## Declarations

**Conflict of interest** The author has no competing interests to declare that are relevant to the content of this article.

**Ethical approval** All work conformed to the legal requirements of the country in which it was carried out.

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