The effects of mud snails on aquatic and terrestrial organisms in paddy fields through development of rice plants

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Chapter 1

General Introduction

The Convention on Biological Diversity defines that biodiversity is the variability among living organisms from all sources, including terrestrial, marine, and other aquatic ecosystems and the ecological complexes. These also raise a question, such as why the biodiversity is important? Here we know that of biodiversity has an important role in the ecosystem function and services (e.g. nutrient cycling and retention, water cycling, maintenance of soil fertility, plant pollination, etc.) (Thébault and Loreau 2006; Power 2010). Following the relationship between biodiversity and ecosystem function, there was a clear evidence that biodiversity has an impact on ecosystem productivity such as the positive effects of plant diversity on decomposer activity and diversity. In addition, the higher plant diversity contributes to lowering plant damage by pest organisms and others (Balvanera et al. 2006, Cardinale et al. 2012). However, much of studies investigating biodiversity is raising concerns about consequences of biodiversity loss for ecosystem functioning or stability (Loreau et al. 2001; Hooper et al. 2005) which depends on species richness, species composition, functional group richness and other factors such as species evenness and genetic diversity (Isbell 2011).

One of the important issues in ecology is to understand the direct and indirect interactions between and/or among organisms involved in ecological communities. To understand the dynamic interactions in a food web which is illustrated by bottom-up and top-down effects has been paid much attention by ecologists, however, our understanding of the dynamic interactions is not complete. “Indirect effect” is a general term referring to a broad variety of species interactions that can occur through chains of direct species interactions, such as predation or interference competition. Still, indirect effects continue to stimulate significant theoretical and experimental works, and comprehensive reviews have discussed their place in modern ecology (Strauss 1991,Wootton 1994). It remains to be seen if indirect effects will persist as a useful unifying concept in the future. Both intraspecific and interspecific competition can occur simultaneously and both can occur via exploitative or interference mechanisms (Vanni et al 2000). Intraspecific competition occurs between individuals of the same species, while interspecifics competition occurs between individuals of two or more species.

In agricultural systems, biodiversity also performs ecosystem services including nutrient recycling, regulation of microclimate, controlling pest organisms and others (Altieri 1999). However, concern is growing about modern agricultural system and, it has become more productive but highly chemical inputs-dependent. Thus modern farming cause several negative effects such as loss of biodiversity, contaminant in the water, soil and air, and also dependency on chemical inputs (Altieri 1995; Pimentel et al. 1992). Considering the advantages and disadvantages of modern agriculture, it is of high importance to reassess all agricultural practices (Li 2001). Developing new concept of environmentally friendly farming (EFF) which has the aim to avoid negative impacts on organisms or environments, has the potential not only to reverse biodiversity loss in farmland but also to have benefits agricultural production through the enhancement of ecosystem services (Bengtsson et al., 2005; Letourneau and Bothwell, 2008).

EFF uses no chemical inputs, making it the most promising method for eliminating the negative effects of conventional farming. In this concept, some farmers use bio-based nutrients from aquatic organisms instead of chemical inputs to improve soil fertility. Several studies have suggested that the use of bio-based nutrients instead of chemical inputs can improve the production by fewer pest insects (Altieri and Nicholls 2003; Butler et al. 2012; Facknath and Lalljee 2005). However, there are few studies to understand the effects of nutrient from aquatic organisms to improve soil fertility, abudance of organisms including aquatic and terrestrial organisms and plant production

Rice fields have been in existence as organized agriculture. It is an ecosystem that sustains not only the people whose staple diet but also a diverse assemblage of plants and organisms including aquatic and terrestrial organisms that have made rice fields their habitats. Moreover, the rice fields offer shelter, food, breeding and nesting grounds and also offer temporary refuge to those animals that are not permanent inhabitants but visit this ecosystem for variety of purposes (Edirisinghe and Bambaradeniya 2010). Several studies to understand the functions of aquatic organisms in paddy fields have focused on positive effects on rice plant development due to bio-based nutrients from aquatic snails (Simpson et al. 1994). Molluscs in particular may have important effects on rice performance by serving as food items (Simpson et al. 1994), and by contributing to nutrient cycling and soil decontamination (Kurihara and Kadowaki 1988). *Cipangopaludina chinensis laeta* (Martens) and *C. japonica* (Martens) (Architaenioglossa: Viviparidae) are two species of mud snails that were previously abundant and commonly found in the paddy fields of east China, Taiwan, Korea, and Japan (Chiu et al. 2002). These mud snails might be key species not only in organizing community structure of aquatic organisms in rice paddies, but also in affecting abundances of aboveground arthropods through development of rice plants. However, there are few studies to understand the effects of the mud snails on aquatic and aboveground communities.

In chapter 1, General introduction, Next, in chapter 2, the study were to examine (1) how the effects of the mud snails on the community structure of aquatic organisms and (2) how the community of aquatic organisms affected on the terrestrial organisms communities through the rice plant development in the paddy field (small scale experiment). Next, in chapter 3, the purpose of this study was to understand (1) how the effects of the mud snails on the community structure of aquatic organisms and their interaction, and (2) how the community of aquatic organisms affected on the terrestrial organisms communities through the rice plant development in the paddy field (large scale experiment). Furthermore, this study also to understand applied aspect from two experiments, how the mud snails influence on rice yields without chemical inputs during rice o farming. Is it possible or not? In addition, chapter 4. General discussion

Chapter 2

Effects of a mud snail *Cipangopaludina chinensis laeta* (Architaenioglossa : Viviparidae) on the abundance of terrestrial arthropods through rice plant development in a paddy field

Introduction

Organisms such as natural enemies and decomposers contribute important ecological services in agro-ecosystems, resulting in decreased pest populations and increased crop yields and rates of nutrient cycling and decomposition (Altieri and Nicholss 1999; Bengtsson et al. 2005; Letourneau and Bothwell 2008). In general, conventional farming methods that rely on pesticides and synthetic fertilizer to achieve high crop productivity have negative impacts on agro-ecosystems and the surrounding environment. These management practices can cause environmental problems such as pest resistance, resurgence, persistence, ground water contamination, and impoverished soil condition (Beilen 2016; Gill and Garg 2014; Hirai 1993; Wilson et al. 2008). Several studies also have reported declines in biodiversity in agro-ecosystems due to pesticides (Garbach et al. 2014) and fertilizer (Tilman et al. 2002; Zhong et al. 2011), including decreases in the abundances of beneficial aboveground arthropods (Natuhara 2013). In order to avoid or minimize these negative effects on agro-ecosystems, organic farming without chemical fertilizer and pesticide has received much attention not only for farmers but also for consumers. However, we have lack essential information regarding the functions of biodiversity in organic agro-ecosystems.

In paddy field ecosystems, organic farming may increase biodiversity of both aquatic and terrestrial communities. For example, Bengtsson et al. (2005) reported a 50% increase in the abundance of organisms in organic versus conventional farming in paddy fields, with greater biodiversity and representation of individual functional groups of arthropods. Rice field ecosystems include both aquatic and terrestrial organisms and support high levels of biodiversity that are essential for the functioning of paddy field ecosystems (Cohen et al. 1994). Paddy fields are temporary, flooded habitats for aquatic insects and other organisms that breed in the shallow water and feed on algae, plankton and other organisms (Mukai et al. 2005). Recent research has begun to reveal the consequences of organic farming for rice plant production (Yadav et al. 2013), but we do not fully understand how organic farming affects the functions of useful organisms involved in the paddy fields.

Several studies of the functions of aquatic organisms in paddy fields have focused on positive effects on rice plant development due to bio-based nutrients from aquatic organisms such as fish (Cagauan 1995; Vromant and Chau 2005) and aquatic snails (Simpson et al. 1994). Molluscs in particular may have important effects on rice performance by serving as food items (Simpson et al. 1994), and by contributing to nutrient cycling and soil decontamination (Kurihara and Kadowaki 1988). *Cipangopaludina chinensis laeta* (Martens) and *C. japonica* (Martens) (Architaenioglossa: Viviparidae) are two species of mud snails that were previously abundant and commonly found in the paddy fields of east China, Taiwan, Korea, and Japan (Chiu et al. 2002). They consume sewage sludge, detritus, periphytic algae, and bacteria (Kurihara and Kadowaki 1988). Management practices in conventional paddy fields have reduced the abundance of these mud snails to low levels (Wilson et al. 2008), but abundances remain high in organic paddy fields (Trisnawati 2015). These mud snails might be key species not only in organizing community structure of aquatic organisms in rice paddies, but also in affecting abundances of aboveground arthropods through development of rice plants. However, there are few studies to understand the effects of the mud snails on aquatic and aboveground communities.

The purpose of this study was to examine (1) how the effects of the mud snails on the community structure of aquatic organisms and (2) how the community of aquatic organisms affected on the terrestrial organisms communities through the rice plant development in the paddy field (cage experiment). We hypothesized that the mud snail would increase aquatic and terrestrial community structure and rice plant development through the year.

Materials and Methods

Experimental site and design

The field study was conducted in 2013 and 2014 at Yamagata University Research Farm, Tsuruoka city, Yamagata prefecture, located in northeastern Japan (38^{o}43`N, 139$^{o}$49`E). During field research, the mean temperature was 21.6 ^{o}C (range 15.4 to 26.0 ^{o}C) and 21.8 ^{o}C (range 16.5 to 25.2 ^{o}C), whereas the rainfall was 245.5 mm and 137.9 mm from May until September in 2013 and 2014, respectively (Japan Meteorological Agency 2013, 2014). The paddy field had been cultivated without chemical input such as fertilizer (inorganic and organic) and pesticides (herbicides, insecticides, and fungicides) since 2007. Experimental plots were established to examine the effect of mud snails on rice plant performance and on abundances of terrestrial and aquatic organisms in the paddy field. Two experimental treatments manipulated the presence of *C. chinensis*: plots with and without mud snails present were created in a completely randomized design with twelve replications each. Each plot was 4 m x 3 m, and ridge plates made of polyvinyl chloride (50cm width x 0.5cm thickness in 2013, 60 cm width x 0.4cm thickness in 2014) were used to enclose plots and prevent movement of the snails among plots. Each plot had a water inlet and an outlet for regulation of water depth. At the inlet and outlet (each rectangular, and 15 cm width x 25 cm height), ridge plates were partly replaced with a mesh (25cm width x 30cm length, mesh size 0.1 cm) to prevent the entry of unwanted organisms such as snails. The distance between adjacent plots was 50 cm and 100 cm in 2013 and 2014, respectively.

Rice plant management

The rice cultivar used in the experiment was Sasanishiki which is commonly grown in this location. The rice plants were cultivated in the small plot paddy field without using chemical inputs such as pesticides and fertilizer (either organic or inorganic) during the experiment. The rice seedlings were transplanted at 30 × 16 cm spacing with one seedling per hill in 250 hills/plot on 1 May in 2013 and 31 May in 2014. After transplanting, all weeds were removed manually at every ten days in 2013 and at every six days in 2014 (i.e. six times in total) before plots were drained in each year. The plots were irrigated by using a water pump from an irrigation ditch. We maintained a water depth of 5cm from transplanting to 14 days after transplanting (DAT) to control transplanting shock for all treatments. After that, a water depth of 5-15cm was maintained according to the condition of rice plants until the period of draining ("*nakaboshi*") which began on 18 July in 2013 and on 27 July in 2014. Water depth in the plots was monitored at intervals of 2-3 days using a wood ruler.

Mud snail

2000 individuals of mud snails were obtained from an irrigation ditch near the study site and maintained in a glass house for two weeks before they were introduced to plots. For plots receiving mud snails, 150 individuals of mud snails were released into each plot after transplanting on 14 May in 2013 and 6 June in 2014, respectively. The number of individuals released was within the density observed in the paddy fields (25.0 ±2.0/m^2 (mean±SE), 10-39/m^2 (range), Trisnawati 2012). Before the introduction, no individuals of the snails were observed in each plot. The mud snails added to plots included both sexes and averaged 27.8 ± 0.2 mm (mean±SE) in shell height (n = 100).

Sampling and identifying of aquatic and terrestrial organisms

In all plots, the abundances of aquatic organisms were determined twice each year, on 22 June and 17 July in 2013, and on 19 June and 14 July in 2014. Abundances of terrestrial arthropods were determined on 1 August and 15 August in 2013, and 15

August and 1 September in 2014. At each date, the aquatic organisms were collected using a 0.5 x 0.2 m acrylic box and a fish net (length 30 cm, diameter 20 cm, 1 mm mesh size) at three randomly selected locations in each plot. We used the fish net to collect aquatic organisms by sweeping six times in the acrylic box area. Whereas for terrestrial organisms, we collected the organisms using a sweeping net (length 150 cm, diameter 40 cm) in the area of 0.5 x 0.5 m by sweeping five times at three selected points in each plot. All samplings were conducted from 7:00 to 11:00 in good weather without rain. The samples were brought back to the laboratory for sorting, identifying and counting. The specimens of aquatic organisms in the water samples or captured terrestrial arthropods were transferred to the plastic bag. The aquatic organisms were identified mainly at the order, family or genus level using a microscope (Olympus SZ-PT, Japan) in the laboratory. All taxa of aquatic organisms were categorized into Functional Feeding Groups (FFGs) based on the references (Merritt and Cummins 1996; Cummins et al. 2005). The FFGs were identified and counted due to the morphological characteristics and behavioral habits of obtaining food. The aquatic organisms that consumed algae and associated materials are referred scrapers such as a freshwater snail (Gastropoda) and mayfly larvae (Ephemeroptera). Gathering collectors like aquatic worms (Oligochaeta) collect fine particulate organic matter (FPOM) from the stream bottom and filtering collectors like midge larvae (Chironomidae) collect FPOM from the water column using a variety of filters. Predators capture and consume alive prey like adults and larvae of predaceous diving beetles (Dytiscidae).

The terrestrial organisms were categorized into three feeding guilds as phytophagous pest insects, natural enemies of the pest insects, and other insects (i.e., all additional insect species which were neither pest insects nor natural enemies) by the references (Moran and Southwood 1982; Settle et al. 1996). The terrestrial arthropods collected from the plots were identified family, genus or species level using available keys using the references (Barrion and Litsinger 1994; Borror and White 1970).

Rice plant performance

Rice plant samples were obtained within plots from randomly selected areas containing ten hills per plot. Rice plant characteristics including leaf color (SPAD value), plant height and tiller number for each of the ten plants sampled from each plot were measured first on 21 June in 2013 and 29 June in 2014, and a second time on 18 July in 2013 and on 26 July in 2014. The uppermost (youngest) expanded leaf on each sampled plant was selected, and a measure of how green it was determined using a SPAD-502 (Konica Minolta Inc., Tokyo, Japan). The SPAD reading was taken from the middle portion of the fully expanded youngest leaf after dividing the leaf into three parts of equal length. In addition, all rice plants from an additional 10 hills were harvested on 15 September in 2013 and 1 October in 2014, and dried in the green house for 10 days before weighing to determine biomass.

Statistical analysis

All statistical analyses were performed with R ver 3.2.3 (R Development Core team 2015). Abundances of aquatic and terrestrial organisms between with and without mud snails added were analyzed by a likelihood ratio (LR) test in a generalized linier model (GLM). We implemented GLM using a Poisson error distribution or for over-dispersion using quasipoisson error distribution with log link function. Deviance from the GLM was analyzed using a chi-squared test. Measurements of rice plant characteristics were analyzed by Welch's *t*-test. Principal components analysis using "stats" package was performed for individual sampling occasions in 2013 and 2014 to characterize differences in the community structure of aquatic and terrestrial organisms between plots with and without mud snail added. The number of mud snails was not included or counted in the data because the presence of mud snails as the treatment. For principal component analysis, the numbers of individuals were transformed using natural logarithms.

Results

Abundance of aquatic organisms

There were no significant differences in the abundance of all aquatic organisms combined in plots with and without mud snails added in either 2013 (Fig. 2.1a; LR test in GLM, June $df = 1$, $\chi^2 = 3.03$, $p = 0.08$; July $df = 1$, $\chi^2 = 1.95$, $p = 0.16$) or in 2014 (June $df = 1$, $\chi^2 = 1.33$, $p = 0.25$; July $df = 1$, $\chi^2 = 1.37$, $p = 0.24$) and between seasons ($df = 1$, $\chi^2 = 1.18$, $p = 0.13$), although the difference in the abundance between years was significant ($df = 1$, $\chi^2 = 119.22$, $p < 0.001$)

There were no significant differences in the abundance of the predators between treatments (i.e., plots with versus without snails added) in 2013 (Fig. 2.1b; June $df = 1$, $\chi^2 = 1.05$, $p = 0.30$; July $df = 1$, $\chi^2 = 4.16$, $p = 0.54$) and in 2014 (June $df = 1$, $\chi^2 = 12.30$, $p = 0.36$; July $df = 1$, $\chi^2 = 17.15$, $p = 0.19$). There were no significant effects of treatment with snails on the abundance of collectors in either year (Fig. 2.2a; 2013, June $df = 1$, $\chi^2 = 1.86$, $p = 0.17$; July $df = 1$, $\chi^2 = 0.02$, $p = 0.90$; 2014, June $df = 1$, $\chi^2 = 2.19$, $p = 0.14$; July $df = 1$, $\chi^2 = 1.45$, $p = 0.23$). Among collectors, tubificid worms and chironomid larvae were especially abundant in all plots. In both 2013 and 2014, there were no significant differences between treatments in the abundance of tubificid worms (2013, June $df = 1$, $\chi^2 = 2.91$, $p = 0.53$; July $df = 1$, $\chi^2 = 0.81$, $p = 0.81$; 2014, June $df = 1$, $\chi^2 = 1.63$, $p = 0.67$; July $df = 1$, $\chi^2 = 1.40$, $p = 0.26$) or chironomid larvae (2013, June $df = 1$, $\chi^2 = 0.11$, $p = 0.83$; July $df = 1$, $\chi^2 = 2.16$, $p = 0.27$; 2014, June $df = 1$, $\chi^2 = 0.23$, $p = 0.60$; July $df = 1$, $\chi^2 = 0.12$,

p = 0.88), although the numbers of two species, tubificids worms and chironomid larvae, tended to be higher in plots to which snails had been added. There were no significant differences in the abundance of scrapers between treatments in 2013 (Fig. 2.2b; June df = 1, χ^2 =1.81, p = 0.18) and in 2014 (June df = 1, χ^2 =0.74, p = 0.39, July df = 1, χ^2 =0.51 p= 0.48).

Community structure of aquatic organisms

As shown by principal components analysis (PCA) the community structures of aquatic organisms in plots with versus without snails added were not significantly different in either June or July in 2013 (Fig. 2.3 a-b) and in 2014 (Fig. 2.4a-b).

Rice plant performance

Table 2.3 shows the characteristics of rice plants in plots with and without snails. The SPAD values of flag leaves did not differ significantly between the treatments in 2013 (Welch's two sample t test, June t = 0.10, p = 0.92; July t = -0.21, p = 0.83) or in July 2014 (July t = -0.44, p = 0.66), but differed significantly in June 2014 (t = 5.59, $p < 0.001$). There were no significant differences in plant height between the treatments in 2013 and 2014 (2013, June t = 0.49, p = 0.63; July t = 0.80, p = 0.43; 2014, June t = -0.78, p = 0.44; July t = -0.61, p = 0.54). However, there were greater numbers of tillers per plant in plots with versus without snails in both 2013 and 2014 (2013, June t = 7.34, $p < 0.001$; July t = 9.89, $p < 0.001$; 2014, June t = 4.07, $p < 0.001$; July t = 3.66, $p < 0.001$). In addition, the plant biomass also tended to be greater in plots with snails, although the difference was significant only in 2014 (2013, September t = -0.87, $p=0.39$; 2014, October t = 3.76, $p < 0.05$).

Abundance of terrestrial organisms

There were significant differences in the abundance of all terrestrial organisms combined as associated with rice plants growing in plots with and without snails in both 2013 (Fig. 2.5a; LR test in GLM, August $df = 1$, $\chi^2 = 188.43$, $p < 0.001$; August $df = 1$, $\chi^2 = 98.12$, $p < 0.001$) and 2014 (August $df = 1$, $\chi^2 = 41.13$, $p < 0.001$; September $df = 1$, $\chi^2 = 45.20$, $p < 0.001$) and between years ($df = 1$, $\chi^2 = 51.13$, $p < 0.001$), although the difference in the abundance did not differ between seasons ($df = 1$, $\chi^2 = 0.13$, $p = 0.08$)

In the case of phytophagous pest insects, the numbers of individuals in plots with snails were higher than those in plots without snails in both 2013 (Fig. 2.5b; August $df = 1$, $\chi^2 = 84.54$, $p < 0.001$; August $df = 1$, $\chi^2 = 54.05$, $p < 0.001$) and 2014 (August $df = 1$, $\chi^2 = 26.86$, $p < 0.001$; September $df = 1$, $\chi^2 = 12.17$, $p < 0.001$). The dominant species in all plots was small brown planthopper, *Laodelphax striatellus* (Fallén) (Delphacidae). The abundance of natural enemies did not differ significantly between the two treatments in either year (Fig. 2.6a; 2013, August $df = 1$, $\chi^2 =0.01$, $p = 0.92$; August $df = 1$, $\chi^2 = 0.81$, $p = 0.37$; 2014, August $df = 1$, $\chi^2 = 1.81$, $p = 0.18$; September $df = 1$, $\chi^2 = 0.66$, $p = 0.42$). There were significant differences in the abundance of other insects (dominated by Chironomidae) between plots with and without snails in both 2013 (Fig. 2.6b; August $df = 1$, $\chi^2 = 105.85$, $p < 0.001$; August $df = 1$, $\chi^2 = 105.82$, $p < 0.001$) and 2014 (August $df = 1$, $\chi^2 = 13.49$, $p < 0.001$; September $df = 1$, $\chi^2 = 45.44$, $p < 0.001$).

Community structure of terrestrial organisms

As shown by principal component analysis, the community structure of terrestrial organisms differed significantly between plots with and without snails in 2013 and 2014. In 2013, the terrestrial organisms that contributed most to the vector of the 1st and 2nd component scores belonged to seven families (Ceratopogonidae, Chironomidae, Delphacidae, Ephydridae, Acrididae, Baetidae, Chrysopidae) on the first sampling occasion, and to four families (Chironomidae, Delphacidae, Dolichopodidae, Empididae) on the second sampling occasion (Fig. 2.7a-b). In 2014, the terrestrial organisms that contributed most to the vector of the 1st and 2nd component scores belonged to four families (Agromyzidae, Dolichopodidae, Chironomidae, Delphacidae) on the first sampling occasion, and to two families (Tetragnathidae and Chironomidae) on the second sampling occasion (Fig. 2.8 a-b).

Discussion

This study clearly showed that the community structure of aquatic organisms in the rice paddy plots was not significantly altered by the addition of mud snails, but this addition did change the community structure of terrestrial organisms as associated with a positive influence on rice plant growth. Hence the mud snails appeared to have bottom up effects on terrestrial organisms resulting from the effects of the mud snails on rice plant development.

In general, interspecific competition for food resources appears the important ecological influence on dynamics of community structure of aquatic organisms (e.g. Hazra and Pal 2014). However, the presence of snails did not significantly affect the abundance of aquatic organisms and in particular the abundance of other scrapers in the present study. This suggests that there was weak interspecific competition for food between the mud snails and other aquatic organisms because food resources (algae) were abundant relative to the low numbers of the feeding group (scrapers) that included the snails in the paddy field throughout the season (Dewi personal observation). As present study was carried out in small enclosures with several limitations, large scale experiment is needed to understand the relationship between the snails and other aquatic organisms since the fact that the enclosures were not completely closed might have made the results more conservative.

Tubificid worms are the dominant aquatic macroinvertebrates in paddy fields (Ito and Hara 2010). High amounts of organic matter and soil nutrients resulted in high density of tubificid worms (Simpson et al. 1993). Nutrients such as nitrate, phosphate and potassium in paddy fields increased the abundance of tubificid worms (Hedge and Sreepada 2014); phosphates and ammonia are produced by mud snails (Underwood et al. 1992). The higher abundance of tubificid worms, the most abundant species present, in plots with snails added in the present study might have been caused by the change in soil nutrient availability resulting from the presence and activity of snails in these plots.

Tiller number is an important agronomic character for rice production (Badshah et al. 2014; Li et al. 2003) which depends strongly on the maximum number of fertile tillers per unit area (Cheng et al. 2015; Halil and Necmi 2005). Tiller number was higher when snails were added to plots, as a result plant biomass tended to be higher in the plot with snails than that without snails. This might come from increased availability of nitrogen associated with snail activity (Baxter et al. 2004; Sabo and Power 2002). Several studies have reported that mud snails accelerated aquatic nutrient recycling in paddy fields by consuming algae and other photosynthetic aquatic biomass and detritus, and then excreting nutrients (Grant et al. 1983). The snails' feeding would thus result in readily decomposing material that enables the plants to directly absorb nitrogen from the soil in the form of nitrate or ammonium ions and phosphate (Xu et al. 2012). Thus, the present study suggests that mud snails play an important role in enhancing rice plant performance through food-web effects that result in nutrient release in the aquatic ecosystem.

In general, plant quantity affects the performance of terrestrial organisms including herbivores, natural enemies and others insects (Kagata et al. 2005; Stiling and Moon 2005; Utsumi and Ohgushi 2009). Previous studies have noted that host plant quantity can directly affect the abundance of phytophagous insects and also can indirectly affect natural enemies as a bottom-up effect (Walker et al. 2008). Our results showed that the snails tended to increase the abundance of phytophagous insects as an indirect effect through the increased biomass of the rice plants, although these bottom-up effects were not detected to affect the abundance of natural enemies. Several previous studies similarly have suggested that host plant quantity did not simultaneously increase the abundance of both phytophagous insects and natural enemies (Perner et al. 2005; Rambo and Faeth 1999), and sometimes had no detectable effect on predator abundance (Koricheva 2000). In the present study, it was clear that the snails' activity increased the biomass of rice plants, which in turn led to an increase in the abundance of phytophagous insects.

Our study showed that the abundance of other above ground insects also was higher in plots with snails, although there was no significant difference between the two treatments in abundance of chironomid larvae (the most abundant of these other above ground insects). Chironomid larvae live in aquatic ecosystems (Pinder 1995), but their adults are terrestrial (Delletre 2005 ; Xu and Wu 1999). Previous studies have revealed that adult chironomids colonize rice fields, and that their populations increase in close association with the development of rice plants (Che Salmah et al. 2000). In addition, the abundance of adult Chironomidae is positively associated with rice plant biomass (Delettre and Morvan 2000). In the present study, the greater number of chironomid adults in plots with snails may have resulted from the greater biomass of rice plants as enhanced by the presence of snails. It therefore appears that the greater abundance of other above ground insects including chironomid adults in plots with snails added likely was resulted from the indirect effect of snails increasing the availability of soil nutrients and increasing plant performance.

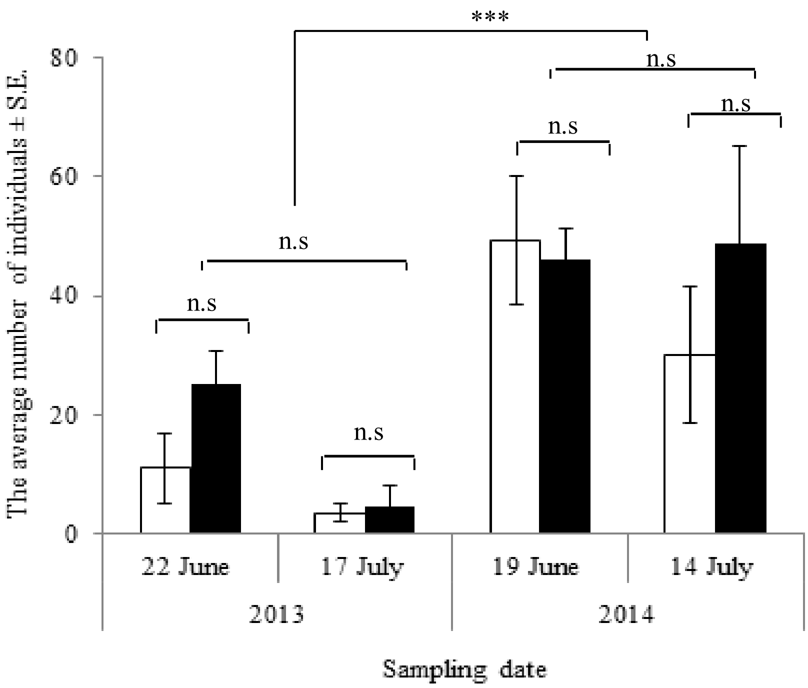
This study suggested that mud snails might play an important role in the paddy field ecosystem, in which they influence the abundance of organisms in terrestrial ecosystems through direct and indirect interactions at different trophic levels. Further investigation using multiple paddy fields is needed to understand the ecological processes underlying the effects of mud snails on arthropods, e.g. aquatic nutrient recycling, in paddy fields since present study was carried out as a small-scale experiment.

Table 2.1. List of aquatic organisms found in the field experiment. The (√) indicates that the aquatic organisms was found in the paddy field with and without mud snail

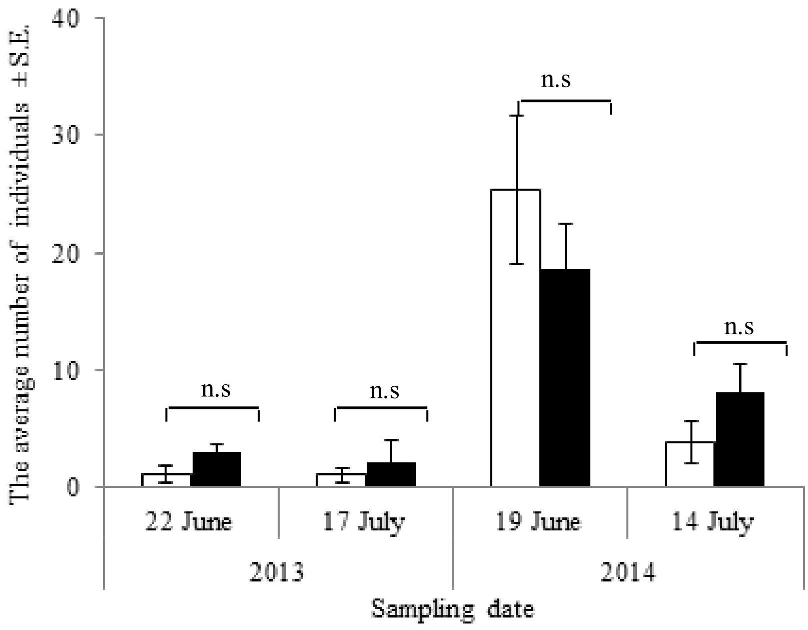
Taxa	Common name	Functional Feeding Group	Mud snails	No Mud Snails
Oligochaetes				
Tubifex tubifex	Sludge worm	C	√	√
Hirudinea				
Glossiphonidae	Leech	P	√	√
Richardsonianidae	Leech	P	√	√
Mollusca				
Sphaeriidae				
Pisidium sp	Pea clams	Sc	√	√
Lymnaeidae				
Radix auricularia japonica	Monoaragai	Sc	√	√
Physidae				
Physa acuta	Bladder snails	Sc	√	√
Coleoptera				
Hydrophilidae				
Berosus punctipennis	Water scavenger beetles	P	√	√
Dytiscidae				
Platambus sp	Predaceous diving beetles	P	√	√
Rhantus suturalis	Predaceous diving beetles	P	√	√
Haliplidae	Crawling water beetles	Sc	√	√
Diptera				
Chironomidae				
Chironomus sp	Chironomid larvae	C	√	√
Ceratopogonidae	Punkies	C	√	√
Ephemeroptera				
Baetidae				
Cleon dipterum	Mayfly larvae	C	√	√
Siphlonuridae				
Siphlonurus sanukensis	Mayfly larvae	C	√	√
Hemiptera				
Gerridae	Water striders	P	√	√
Notonectidae	Backwimmer	P	√	√
Corixidae	Water boatmen	P	√	√
Loach		P	√	√
Amphibi				
Rana japonica	Tadpole	C	√	√

Abbreviation : P (Predator), C (Collectors including collectors gatherers and filtering collectors, Sc (Scrapers),

(a) Total



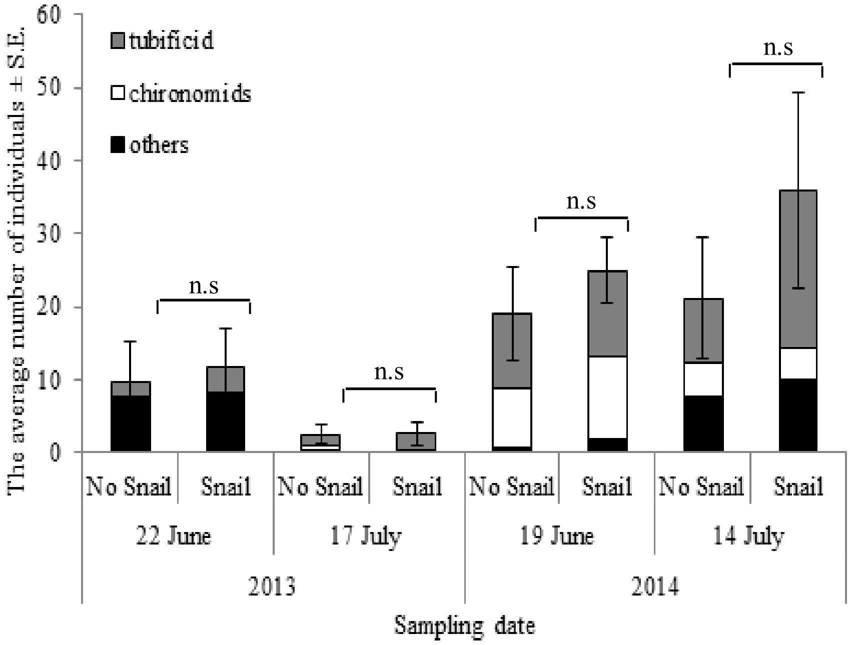
(b) Predator



□ No Snails ■ Snails

Fig. 2.1 The average number (±SE) of individuals at treatments with and without mud snails during two years. (a) Total number of aquatic organisms and (b) Predator. Data were analyzed using the likelihood ratio (LR) test in generalized linier models (GLMs) with poisson and quasipoisson error distribution. (***p<0.001, ns: not significant)

(a) Collector



(b) Scraper

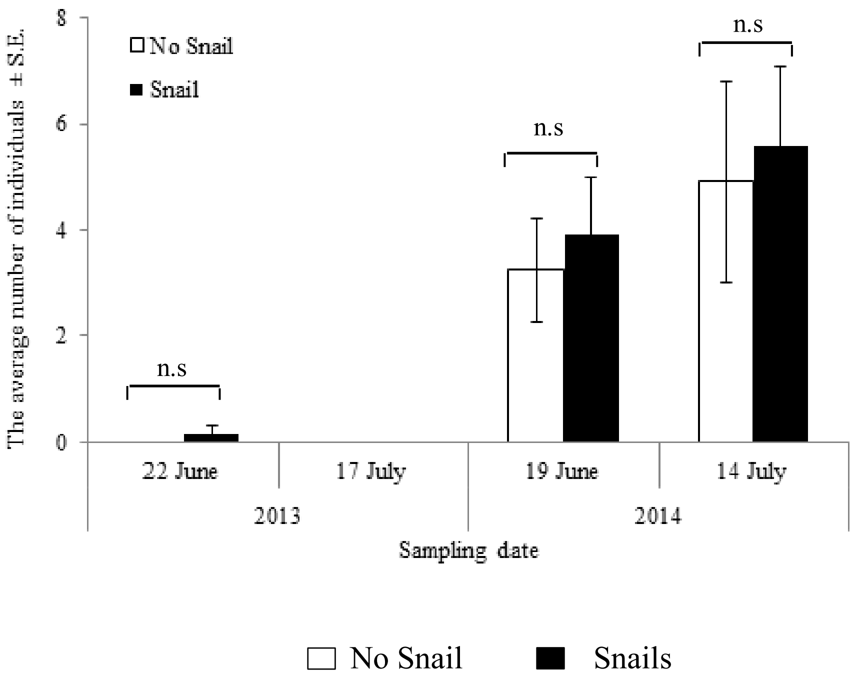
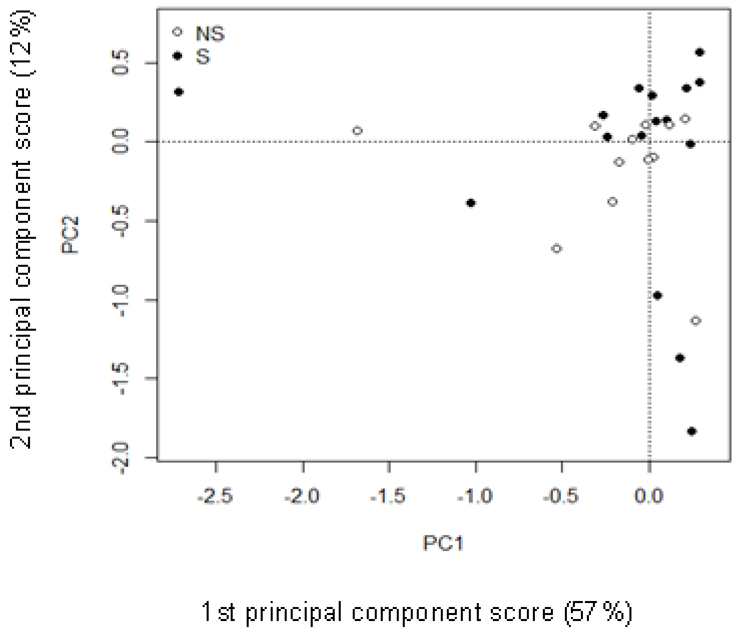


Fig. 2.2 The average number (±SE) of individuals at treatments with and without mud snails during two years. (a) Collector (tubificids, chironomids, others), and (b) Scraper. Data were analyzed using the likelihood ratio (LR) test in generalized linier models (GLMs) with poisson and quasipoisson error distribution. (ns: not significant)

First Observation in 2013



(b) Second Observation in 2013

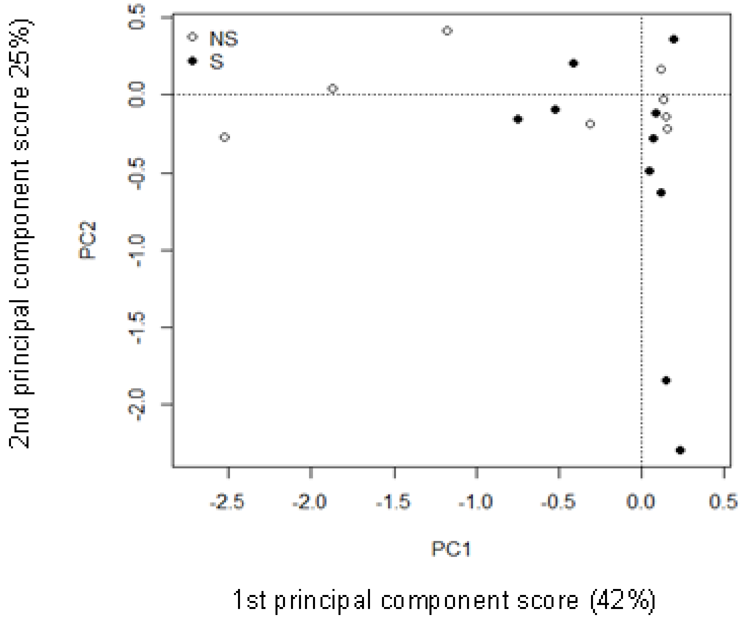
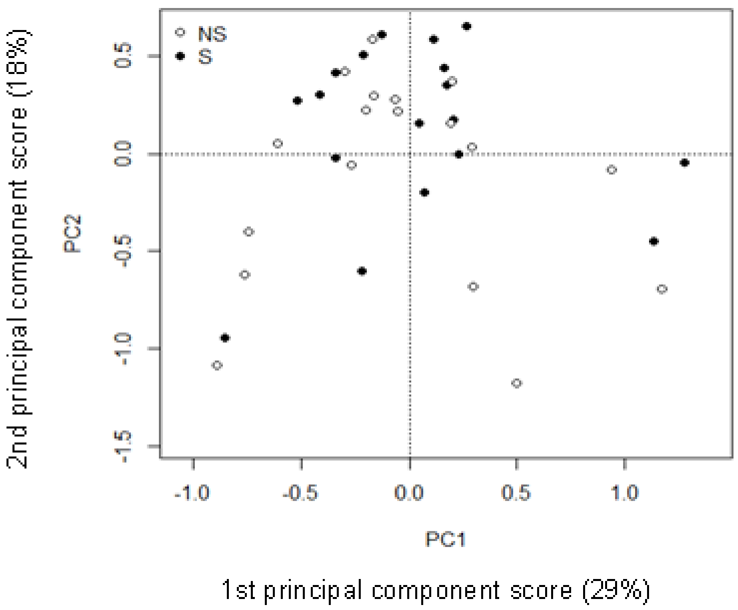


Fig. 2.3 Principal Components for community structure of aquatic organisms during two observations before the drained season : (a) first observation in 2013, and (b) second observation in 2013. Black circles represent the data at treatment with snails and white circles represent the data without snails. Numerals with % indicate the percentage contributed by the 1st principal component analysis or 2nd one.

(a) First Observation in 2014



(b) Second Observation in 2014

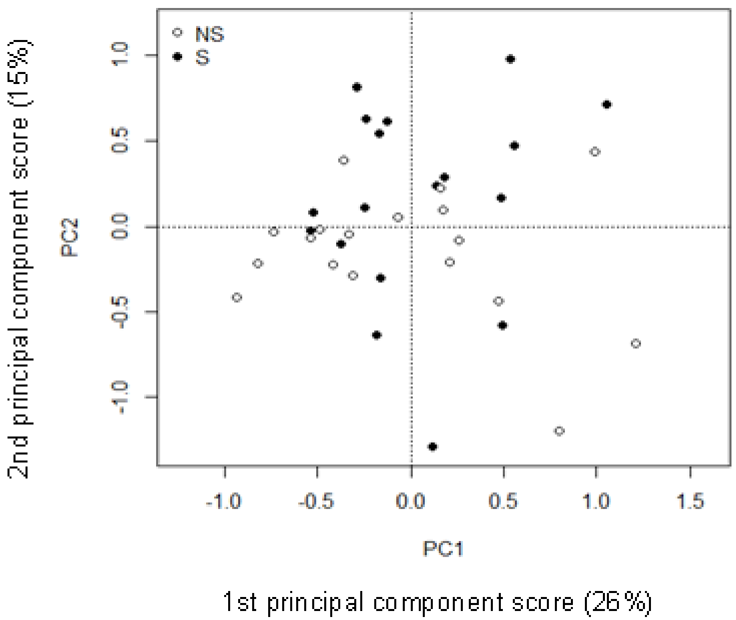


Fig. 2.4 Principal Components for community structure of aquatic organisms during two observations before the drained season : (a) first observation in 2014 and (b) second observation in 2014. Black circles represent the data at treatment with snails and white circles represent the data without snails. Numerals with % indicate the percentage contributed by the 1st principal component analysis or 2nd one.

Table 2.2. List of terrestrial arthropods found in the study area. The (√) indicates that the terrestrial arthropod was found in paddy field with and without mud snail treatment

Taxa	Feeding guilds	Mud snails	No Mud Snails
Hemiptera			
Delphacidae	Herbivore	√	√
Nabidae	Herbivore		√
Coreidae	Herbivore		√
Alydidae	Herbivore		√
Meloidae	Predator	√	
Miridae	Herbivore	√	√
Pentatomiidae	Herbivore	√	√
Homoptera			
Coccidae	Herbivore	√	√
Thripidae	Herbivore		√
Aphididae	Herbivore	√	√
Diptera		√	√
Pomaceae	Herbivore	√	√
Ceratopogonidae	Other insect	√	√
Chironomidae	Other insect	√	√
Tipulidae	Other insect	√	√
Mymariidae	Natural enemies	√	√
Syrphidae	Natural enemies	√	√
Dolicopodidae	Natural enemies	√	√
Simuliidae	Other insect		
Chamamyiidae	Other insect	√	
Milichidae	Other insect	√	
Mycetophilidae	Other insect	√	√
Chloropidae	Other insect	√	√
Agromyzidae	Natural enemies	√	√
Cecidomyiidae	Other insect	√	√
Drosophilidae	Other insect	√	√
Acroceriidae	Natural enemies	√	√
Orthoptera		√	√
Acrididae	Herbivores	√	√
Tettigonidae	Natural enemies	√	
Neuroptera			
Chrysopidae	Natural enemies	√	√
Hymenoptera			
Torymiidae	Natural enemies	√	√
Braconidae	Natural enemies	√	√
Chalcidoidae	Natural enemies	√	√
Eulopidae	Natural enemies	√	√
Eupelmidae	Natural enemies	√	√
Scelionidae	Natural enemies	√	
Trichogrammatidae	Natural enemies	√	√
Platygasteridae	Natural enemies	√	√
Cynipidae	Herbivore	√	√
Pteromaliidae	Natural enemies		√
Coleoptera			
Curculionidae	Herbivore	√	√
Coccinelidae	Predator	√	√
Spider			
Tetragnathidae	Predator	√	√
Thomisidae	Predator	√	√
Lynipidae	Predator	√	√
Spider mites	Predator	√	√

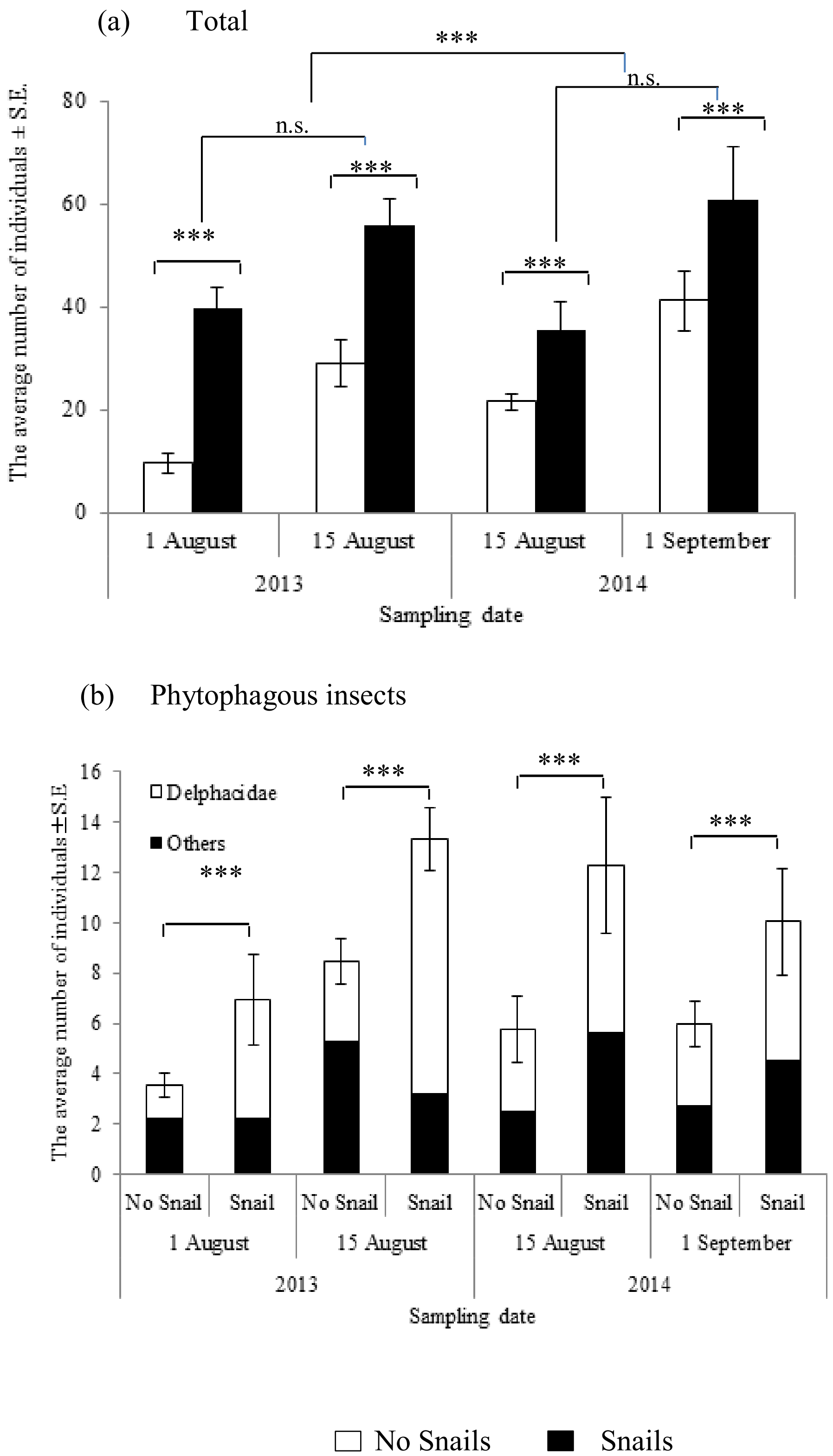
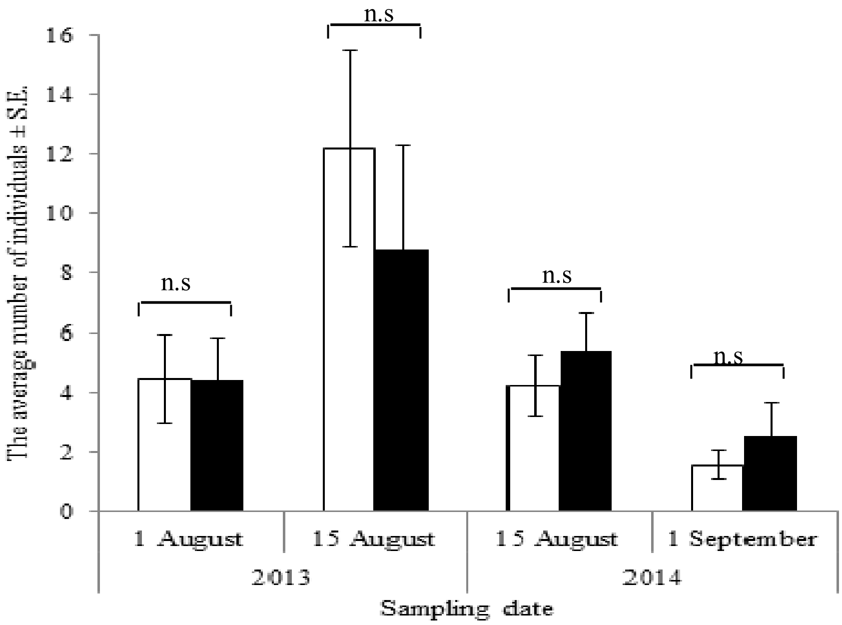
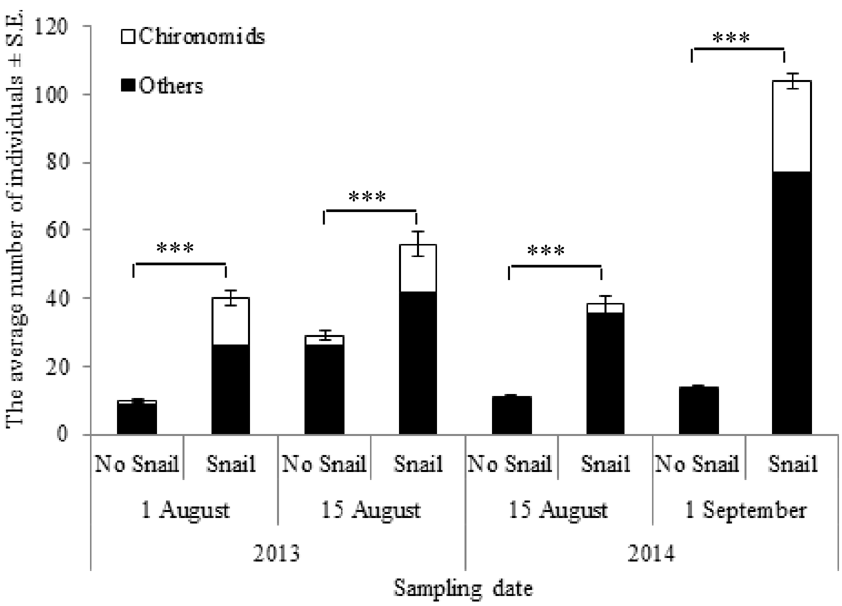


Fig. 2.5 The average number (±SE) of individuals at treatments with and without mud snails during two years. (a) Total number of terrestrial organisms, and (b) Phytophagous insects (Delphacidae and others). Data were analyzed using the likelihood ratio (LR) test in generalized linier models (GLMs) with poisson and quasipoisson error distribution. (****p*<0.01, ns: not significant)

(a) Natural enemies



(b) Other insects



☐ No Snails ■ Snails

Fig. 2.6 The average number (±SE) of individuals at treatments with and without mud snails during two years. (a) Natural enemies, and (b) Other insects (Chironomids and others). Data were analyzed using the likelihood ratio (LR) test in generalized linier models (GLMs) with poisson and quasipoisson error distribution. (****p*<0.01, ns: not significant)

(a) First observation in 2013 **(b)** Second Observation in 2013

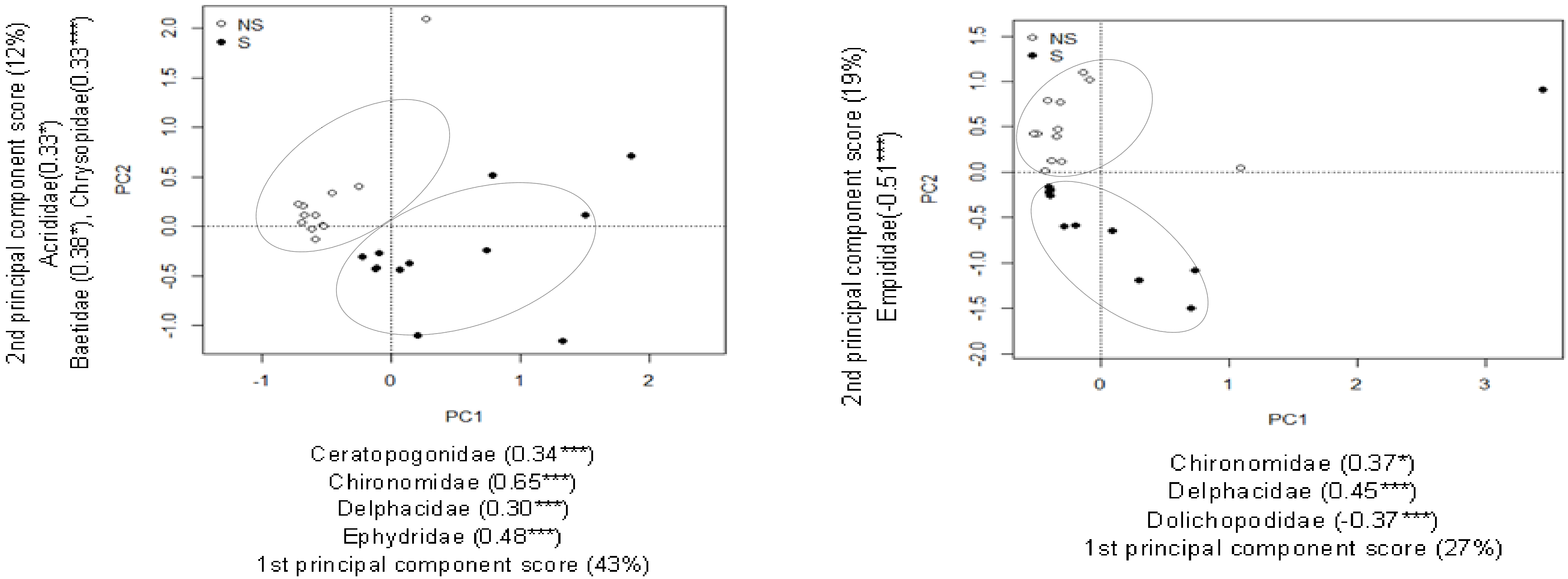
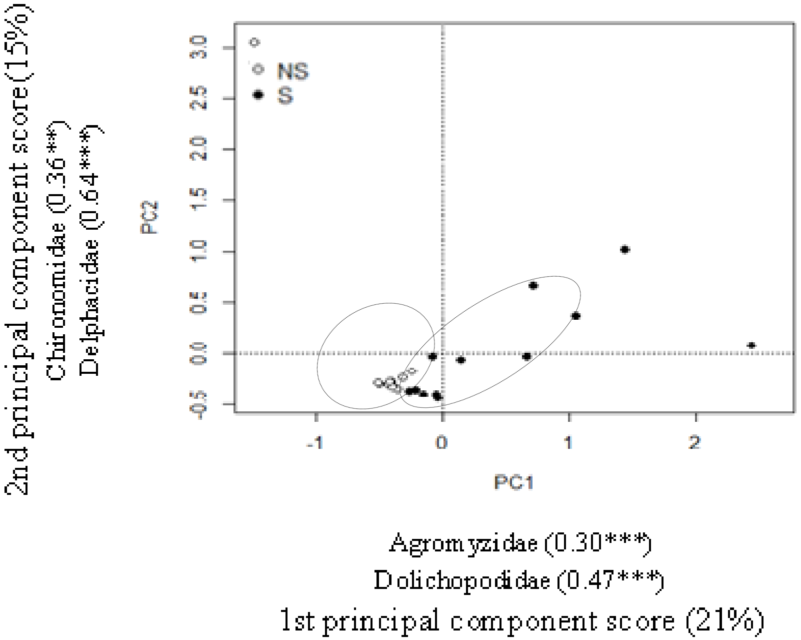


Fig. 2.7 Principal Components for community structure of terrestrial organisms in paddy fields during two observations: (a) first observation in 2013, and (b) second observation in 2013. Black circles represent the data at treatment with snails and white circles represent the data without snails. Numerals with % indicate the percentage contributed by the 1st principal component analysis or 2nd one. Names of aquatic macro-invertebrates refer to those that contributed to the respective vectors. Numerals in parentheses indicate factor loadings (* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$).

(a) First observation in 2014



(b) Second Observation in 2014

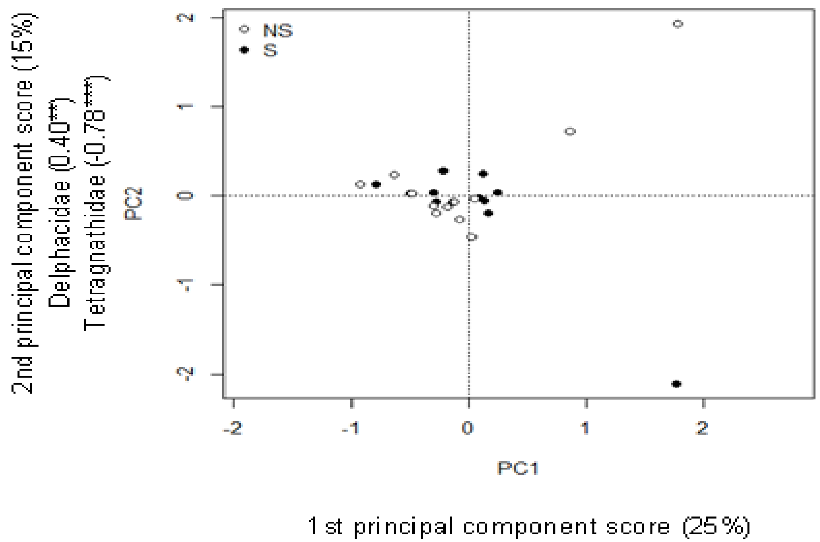


Fig. 2.8 Principal Components for community structure of terrestrial organisms in paddy fields during two observations: (a) first observation in 2014, and (b) second observation in 2014. Black circles represent the data at treatment with snails and white circles represent the data without snails. Numerals with % indicate the percentage contributed by the 1st principal component analysis or 2nd one. Names of aquatic macro-invertebrates refer to those that contributed to the respective vectors. Numerals in parentheses indicate factor loadings (* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$).

Table 2.3 Leaf colour, plant height, tiller number, and rice plant biomass/hill in the plots with and without snails in 2013 and 2014

Years	Treatment	Leaf colour (SPAD Value)		Plant height (cm)		Tiller number per hill		Plant biomass (dry weight) (g)
		June	July	June	July	June	July	
2013	No Snails	(38.4 ± 0.3)a	(39.2 ± 0.2)a	(38.7 ± 0.3)a	(59.0 ± 0.7)a	(4.5 ± 0.04)a	(10.9 ± 0.1)a	(658.0 ± 37.5)a
	Snails	(37.9 ± 0.3)a	(39.2± 0.2)a	(38.8 ± 0.3)a	(59.8 ± 0.6)a	(5.6 ± 0.1)b	(13.3 ± 0.1)b	(672.5 ± 30.3)a
2014	No Snails	(38.8 ± 0.2)a	(39.8 ± 0.2)a	(34.3 ± 0.4)a	(58.7 ±0.3)a	(5.5 ± 0.1)a	(21.8 ±0.4)a	(854.0 ± 28.9)a
	Snails	(40.3± 0.2)b	(39.7 ± 0.2)a	(33.9 ± 0.4)a	(58.5 ± 0.3)a	(6.4 ± 0.2)b	(24.0 ± 0.5)b	(961.0 ± 23.5)b

Values (mean ± SE) within each column followed by the same letters are not significantly different between the treatments by Welch`s t test (p>0.05). The data of plant biomass was obtained from sampling on September and October in 2013 and 2014, respectively

Chapter 3

Effects of a mud snail *Cipangopaludina chinensis laeta* (Architaenioglossa : Viviparidae) on the abundance of aquatic organisms, terrestrial arthropods and rice plant development in a paddy field (large scale experiment)

Introduction

Food web is a process in a biological communities that includes the interaction between producers and consumers in the trophic level. Moreover, energy flows that occur in the food chain is the transfer of energy between living organisms and their environment (Hunter 2001; Peter et al. 2005). The dynamics of subsidies in the ecosystems are also influenced by top-down and bottom-up effect which is regulated by species trophic interactions (Larsen et al. 2016). Thus, the diversity of the species involved in the interaction among trophic levels affect processes in the ecosystem, e.g. the transfer of subsidies (Romero and Srivastava 2010). In fact, detritus feeder like some of the larvae are important prey for aquatic predators, whereas the adult stages are important consumers in the terrestrial ecosystems (Knight et al 2005).

Aquatic organisms inhabit the soil-floodwater ecosystem of paddy fields and are an important component of paddy fields (Simpson and Roger, 1993), which contribute to nutrient cycling from their activities. Grazers and detritivores including microcrustaceans, aquatic insects, gastropods, oligochaetes perform important roles in the decomposition of the photosynthetic aquatic biomass, which develops in ricefield ecosystems. Moreover, tubificid worms have an important role in ensuring the translocation of organic matter that accumulates in the detritus layer at the soil-water interface. Other organisms such as aquatic invertebrates also contribute to nutrient cycling and release native minerals of the soil from their activities (Grant, Roger, and Watanabe, 1986). Covich et al. (1999)

suggested that some species have a large impact on food-web dynamics and provide essential ecosystem services. Thus, aquatic and soil organisms are important components of ricefield fertility (Roger, Grant, Reddy, and Watanabe, 1987; Roger and Kurihara, 1988).

In fact, conventional farming causes several negative effects such as reduce of biodiversity, contaminant in the air, soil and water and also high dependency on chemical inputs (Altieri 1995; Pimentel et al. 1992). Another consequences resulting from long journey of chemical application on soil are toxic chemicals contamination of surface and groundwaters. In other cases, the particulary troblesome is excess of nitrat derived from chemical inputs including fertilizer and pesticides aimed at harmful insects, weeds, and pathogenic fungi (Jon 1999). In fact, there are several ways to minimize the use of chemical inputs for pest, weed, or disease control strategies in some region (Hauptili et al. 1990). Thus, the critical point to minimize the use of chemical inputs to agroecosystems was selected as one of the important factors to be presented.

Agricultural environments are essential for the maintenance of the world`s biological diversity and their agronomic sustainability (Bambaradeniya et al. 2004; Matson et al. 1997; Swift and Anderson 1994). In rice field ecosystem, a variety of aquatic organisms have colonized and they are considered as ecosystem engineers, as to play a vital role in food web dynamics (Bambaradeniya 2000). Several studies of the function of aquatic organisms in the paddy fields have concentrated on positive effects on rice plant development due to bio-based nutrients from aquatic snails (Simpson et al. 1994). In previoust studies, the mud snail *C. chinensis* gave positive influence on community structure of terrestrial organisms through rice plant development (Dewi 2016). In this experiment, we would like to undertand the consequences of mud snail influence on community structure of aquatic organisms, terrestrial arthropods and rice plant development in large scale experiment because there is bottom up mechanisms in their interactions.

The purpose of this study was to understand how the mud snaila influence on (1) the community structure of aquatic organisms, (2) the community structure of terrestrial organisms, and (3) the rice plant development in the paddy field. Thus, I hypothesized that the addition of mud snail would more clearly influence on community structure of aquatic organisms in the paddy fields, in particular,negatively on some species in similar guilds. The mud snails also would positively influence on rice plat growth and indirectly positively influence on phytophagous insects and natural enemies.

Materials and Methods

Experimental site and design

The field study was conducted in 2013, 2014 and 2015 at Matsugaoka Farm, Tsuruoka city, Yamagata prefecture, located in northeastern Japan (38°43`N, 139°49`E). During field research, the mean temperature was 21.6 °C (range 15.4 to 26.0 °C), 21.8 °C (range 16.5 to 25.2 °C) and 21.8 °C (range 18.1 to 20.5 °C), whereas the rainfall was 245.5 mm, 137.9 mm, and 99.6 mm from May to September in 2013, 2014 and 2015, respectively (Japan Meteorological Agency 2013, 2014 and 2015). The Japonica type rice plants (*Oryza sativa*) were cultivated in the paddy field without chemical input such as fertilizer (inorganic and organic) and pesticides (herbicides, insecticides, and fungicides) since 2012. Experimental fields were established to examine the effect of mud snails on abundances of terrestrial and aquatic organisms through rice plant performance in the paddy field. Two experimental treatments manipulated the presence of *C. chinensis* were made at the fields with and without mud snails with three replications each. Each field was 100 m x 5 m, and ridge plates made of polyvinyl chloride (50cm width x 0.5cm thickness) were used to separate a field into two subfields because of preventing from immigration of the snails from the field with snails treatment. Each field had a water inlet and an outlet for irrigation.

Rice plant management

The rice cultivar used in the experiment was Koshihikari which is a popular variety of rice cultivated in Japan as well as Australia and the United States. The rice plants were cultivated in the paddy field without using chemical inputs such as pesticides and fertilizer during the experiment. Rice seedlings were transplanted at 30 × 15 cm spacing with 5-6 seedlings per hill on 26 May in 2013, 17 May in 2014 and 18 May in 2015. After transplanting, all weeds were removed by a weeding machine at every six days in 2013, 2014 and 2015 (i.e. six times in total) before fields were drained in each year. The fields were irrigated directly from an irrigation ditch. We maintained irrigation by the condition of rice plants until the period of draining ("*nakaboshi*") which began on 15 July in 2013, 20 July in 2014 and on 25 July in 2015.

Mud snail

3000 individuals of mud snails were obtained from an irrigation ditch near the Takasaka farm and maintained in a glass house for one weeks before they were introduced to the paddy field. For the fields receiving mud snails, 1000 individuals of mud snails were released into each field after transplanting on 9 June in 2013, 23 May in 2014 and 24 May in 2015, respectively. Before the introduction, no individuals of the snails were observed in each field. The mud snails added to the field included both sexes and averaged 25.84 ± 0.25 mm (mean±SE) in shell height (n = 150).

Sampling and identifying of aquatic and terrestrial organisms

In all fields, the abundances of aquatic organisms were determined four times each year, on 16 June, 25 June, 5 July and 14 July in 2013, on 4 June, 21 June, 1 July and 19 July in 2014, and on 5 June, 23 June, 6 July, and 23 July in 2015. Abundances of terrestrial arthropods were determined on 20 July, 4 August, 19 August, and 3 September in 2013, on 1 August, 25 August, 8 September, and 22 September in 2014, on 3 August, 28 August, 9 September, and 21 September in 2015. At each sampling date, water and aquatic organisms were collected using 100-ml beaker and a fish net (length 30 cm, diameter 20 cm, 1 mm mesh size) in a 0.5 x 0.2 m acrylic box. The sampling was done at 15 randomly selected sites at each field and total sampling was 90. The fish net was used to collect aquatic organisms by six times sweeping in the acrylic box area. For terrestrial organisms, the organisms were collected using a sweeping net (length 150 cm, diameter 40 cm) in the area of 1 m x 3 m by ten times sweeping at 5 randomly selected sites. All samplings were conducted from 6:00 to 10:00 in good weather without rain. The samples were brought back to the laboratory for sorting, identifying and counting. The specimens of aquatic organisms in the water samples and captured terrestrial arthropods were transferred to the plastic bag. The aquatic organisms were identified mainly at the order, family or genus level using a microscope (Olympus SZ-PT, Japan) in the laboratory. All taxa of aquatic organisms were categorized into Functional Feeding Groups (FFGs) based on the references (Merritt and Cummins 1996; Cummins et al. 2005). The FFGs were identified and counted due to the morphological characteristics and behavioral habits of obtaining food. The aquatic organisms that consumed algae and associated materials were referred scrapers such as a freshwater snail (Gastropoda) and mayfly larvae (Ephemeroptera). Gathering collectors like aquatic worms (Oligochaeta) collect fine particulate organic matter (FPOM) from the stream bottom and filtering collectors like midge larvae (Chironomidae) collect FPOM from the water column using a variety of filters. Predators capture and consume alive prey like adults and larvae of predaceous diving beetles (Dytiscidae).

The terrestrial organisms were seperated into three feeding guilds as phytophagous pest insects, natural enemies of the pest insects, and other insects (i.e., all additional insect species which were neither pest insects nor natural enemies) by the references (Moran and Southwood 1982; Settle et al. 1996). The terrestrial arthropods collected from the plots were identified family, genus or species level using available keys using the references (Barrion and Litsinger 1994; Borror and White 1970).

Rice plant performance

Rice plant samples were obtained within fields from randomly selected areas containing 50 hills per field. Rice plant characteristics including leaf color (SPAD value), plant height and tiller number for each of the 50 plants sampled from each field were measured first on 17 June, 23 July, 3 August, 3 September in 2013, on 4 June, 23 July, 4 August, 20 September in 2014, and on 18 June, 20 July, 4 August, 15 September in 2015. The uppermost (youngest) expanded leaf on each sampled plant was selected, and a measure of the degree of greenness it was determined using a SPAD-502 (Konica Minolta Inc., Tokyo, Japan). The SPAD reading was taken from the middle portion of the fully expanded youngest leaf after dividing the leaf into three parts of equal length. In addition, all rice plants from an additional 100 hills at selected sites were harvested on 17 September in 2013, on 25 September in 2014, and 5 October in 2015, and dried in a green house for 10 days before weighing to determine biomass.

Statistical analysis

All statistical analyses were performed with R ver 3.2.3 (R Development Core team 2015). Abundances of aquatic and terrestrial organisms between with and without mud snails added were analyzed by a likelihood ratio (LR) test in a generalized linier model (GLM). We implemented GLM using a Poisson error distribution or for over-dispersion using quasipoisson error distribution with log link function. Deviance from the GLM was analyzed using a chi-squared test. Measurements of rice plant characteristics were analyzed by Welch's *t*-test. Principal components analysis using "stats" package was performed for individual sampling occasions in 2013, 2014, 2015 to characterize differences in the community structure of aquatic and terrestrial organisms between plots with and without mud snails added. The number of mud snails was not included or counted in the data because of the presence of mud snails as the treatment. For principal component analysis, the numbers of individuals were transformed using natural logarithms.

Results

Abundance of aquatic organisms

There were ten taxonomical groups of aquatic macro-invertebrates as order levels and they were identified mainly at the family level in the study fields (Table 1). Based on the functional feeding group, the aquatic macro-invertebrates found in the study fields were categorized into three groups such as predators, collectors including filtering collectors and gatherers, and scrapers. The total abundance of aquatic macro-invertebrates tended to be higher in snail treatment and the trend was similar by the year, although the total abundance of aquatic organisms in both of treatment were decreased in 2015 (Fig. 3.1). There were significant differences in the abundance of all aquatic organisms in fields with and without mud snails in 2013, 2014 and 2015 (LR test in GLM, 2013 $df = 1$, $\chi^2 = 10.61$, $p < 0.01$; 2014 $df = 1$, $\chi^2 = 78.73$, $p<0.05$; 2015 $df = 1$, $\chi^2 = 381.53$, $p<0.001$). Collectors were most abundant group in aquatic community both in the treatments with and without mud snails. However, the total abundance of collectors was higher in the snail treatment during three years. There were significant differences in the abundance of Dystiscidae and Hydrophilidae between the treatments in 2013, but not for red mites (Fig. 3.2a; 2013, Dytiscidae $df = 1$, $\chi^2 = 58.20$, $p< 0.01$; Hydrophilidae $df = 1$, $\chi^2 = 60.16$, Red mites $p< 0.01$; $df=1$, $\chi^2 = 10.2$, $p=0.42$) . In 2014 and 2015, there were no significantly differences in abundance of predators between the two treatments except leech (Fig. 3.2b-c; 2014, Dytiscidae $df = 1$, $\chi^2 = 4.20$, $p = 0.25$; Others $df = 1$, $\chi^2 = 2.19$, $p = 0.57$; 2015, Dytiscidae $df = 1$, $\chi^2 = 3.0$, $p = 0.26$; Ceratopogonidae, $df = 1$, $\chi^2 = 2.50$, $p = 0.06$; Leech, $df = 1$, $\chi^2 = 111.20$, $p < 0.01$;Others, $df = 1$, $\chi^2 = 1.01$, $p = 0.10$). There were significant effects of treatment with snails on the abundance of collectors in 2014 and 2015, but not in 2013 (Fig. 3.3a-c; 2013 Ostracoda, $df = 1$, $\chi^2 = 1.56$, $p = 0.19$; Oligochaetes $df = 1$, $\chi^2 = 0.01$, $p = 0.87$; 2014, Ostracoda $df = 1$, $\chi^2 = 72.11$, $p < 0.05$; Cyclopoida $df = 1$, $\chi^2 = 81.55$, $p<0.05$; 2015, Ostracoda, $df = 1$, $\chi^2 = 121.55$, $p< 0.01$;

Copepoda, *df* = 1, χ^2 =91.0, p< 0.01; Tubificidae, *df* = 1, χ^2 =181.22, p< 0.001; Others, *df* = 1, χ^2 =1.55, p>0.05). There was significantly difference in the abundance of scrapers between the treatments but not in 2014 (Fig 3.4a-b, 2013, Cladocera *df* = 1, χ^2 =34.91, p = 0.05; 2014, Cladocera *df* = 1, χ^2 =1.73, p>0.05; Physidae *df* = 1, χ^2 =1.50, p>0.05).

Community structure of aquatic organisms

Principal component analysis (PCA) in 2013 showed that the community structure of aquatic organisms was significantly different between with and without snails in 2013. It also depicted the aquatic organisms that contributed to the vector of the 1st component scores or 2nd ones, showing that Cyclopoida, Oligochaetes, and Cladocera characterized the aquatic community structure (Fig. 3.5a). In addition, PCA analysis in 2014 and 2015 also resulted that there were significantly differences in community structure of aquatic organisms between the two treatments (Fig. 3.5b-c). It also depicted the aquatic organisms that contributed to the vector of the 1st component scores or 2nd ones, showing that Ostracoda, Oligochaetes, and Copepoda characterized the aquatic community structure in 2014, and Ostracoda, Oligochaetes, and Cyclopoida in 2015 (Fig. 3.5c).

Rice plant performance

Table 2 shows the characteristics of rice plants in plots with and without snails. There were no significant differences in plant height between the treatments in 2015 and 2014 except in 2013 (2013, t = 3.5864, p <0.001; 2014, t = -1. 87, p>0.05; 2015, t = -0.26, p>0.05). However, there was the greater number of tillers per hill in fields with snails in 2013, 2014 and 2015 (2013, t = -4.19, p < 0.001; 2014, t = -1.91, p < 0.001; 2015, t = -2.69, p < 0.001). The plant biomass also tended to be higher in fields with snails, although the difference was significant only in 2014 and 2015 (2013, t = -0.77, p>0.05;

2014, t = -2.39, $p < 0.05$; 2015, t = -2.06, $p<0.05$). In addition, brown rice yields were significantly higher in the field with snails in 2014 and 2015 but not in 2013 (2013, t = -0.77, $p=0.44$; 2014, t = -2.09, $p < 0.05$; 2015, t = -2.18, $p<0.05$). The panicle number showed significantly difference between the treatments in 2014 and 2015 (Fig. 3.8; 2014, $p <0.01$; 2015, $p<0.01$) but no significant difference on length of panicle in 2015 ($p>0.05$). The SPAD values of flag leaves showed significantly difference between the treatments in 2013, 2014, and 2015 (Fig. 3.9; 2013, t = -3.00, $p <0.01$; 2014 t = -5.33, $p<0.01$); 2015 (t = -2.26, $p<0.05$).

Abundance of terrestrial organisms

Forty nine taxonomical groups of terrestrial organisms were shown in Table 3-3 and they were identified mainly at the family levels. Based on the feeding guilds, the terrestrial organisms founded in the study fields were categorized into three groups, herbivores, natural enemies and neutral insects. There were significant differences in the abundance of all terrestrial organisms in fields with and without mud snails in 2013 (Fig. 3-10; 2013, LR test in GLM, 2013 $df = 1$, $\chi^2 =93.04$, $p < 0.001$), but no significant differences in 2014 (2014, $df = 1$, $\chi^2 =2.03$, $p=0.67$), although the difference in the abundance also shown in 2015 (2015, $df = 1$, $\chi^2 = 86.11$, $p < 0.001$). The total abundance of terrestrial organisms was higher in the snail treatment and the trend was similar for three years and alo other insects were most abundant group in terrestrial community both in the treatments with and without mud snails (Fig. 3.10).

The abundance of herbivores tended to be higher in the field with snails, although the differences were not significant for given species and for some year (Fig. 3.11a-c; 2013, LR test in GLM, Delphacidae $df = 1$, $\chi^2 = 88.43$, $p < 0.001$; Acrididae $df = 1$, $\chi^2 = 118.12$, $p < 0.001$; Ephydridae $df = 1$, $\chi^2 = 41.13$, $p < 0.01$; 2014, Delphacidae $df = 1$, $\chi^2 = 2.5$, $p>0.05$; Ephydridae, $df = 1$, $\chi^2 = 61.13$, $p < 0.01$; Others $df = 1$, $\chi^2 = 1.05$,

p>0.05; 2015, Delphacidae df = 1, χ^2 = 42.5, p<0.05; Ephydridae, df = 1, χ^2 = 1.60, p>0.05; Aphididae, df = 1, χ^2 = 2.7, p=0.21; Others, df = 1, χ^2 = 3.0, p>0.05). The dominant species was Delphacidae, Acrididae and Ephydridae. The abundance of natural enemies tended to be higher in snail treatment, although significantly differed between the two treatments in either year except Braconidae in 2013 and 2015 (Fig. 3.12a-c; 2013, Braconidae df = 1, χ^2 =80.01, p<0.05; Dolicopodidae df = 1, χ^2 = 0.71, p > 0.05; Platygasteridae df = 1, χ^2 = 0.77, p > 0.05; Others df = 1, χ^2 = 0.80, p > 0.05; 2014, Eupelmide df = 1, χ^2 = 1.81, p > 0.05; Others df = 1, χ^2 = 0.67, p > 0.05; 2015, Braconidae df = 1, χ^2 = 77.71, p<0.05). In case of other/neutral insects, there were similar trends in each year at which the treatment with snails tended to be higher and dominated by Chironomidae, Ceratopognidae and Plecoptera, although there were no significant differences in the abundance between the treatments except in 2015 (Fig. 3.13a-c; 2013, Ceratopogonidae df = 1, χ^2 = 1.85, p > 0.05; Chironomidae df = 1, χ^2 = 1.2 , p > 0.05; 2014, Ceratopogonidae df = 1, χ^2 = 2.49, p > 0.05; Chironomidae df = 1, χ^2 = 1.44, p=0.34; Others df = 1, χ^2 = 0.72, p > 0.05; 2015, Ceratopogonidae df = 1, χ^2 = 0.61, p > 0.05; Chironomidae df = 1, χ^2 = 90.71, p <0.001; Plecoptera, df = 1, χ^2 = 161.0, $p<0.001$; Others df = 1, χ^2 = 10.1, p > 0.05).

Community structure of terrestrial organisms

As shown by principal component analysis (PCA), the community structure of terrestrial organisms differed significantly between the fields with and without snails in 2013 (Fig. 3-14) and 2015 (Fig. 3-16) except in 2014 (Fig. 3-15). In 2013, the terrestrial organisms that contributed most to the vector of the 1st and 2nd component scores belonged to three families Ceratopogonidae, Chironomidae, Delphacidae (Fig. 3.14). However, PCA analysis in 2014 resulted that community structure of terrestrial organisms between the treatments with and without snails was no significant difference

(Fig. 3.15) In 2015, the terrestrial organisms that contributed most to the vector of the 1st and 2nd component scores also belonged to three families Chironomidae, Ceratopogonidae and, Delphacidae on the (Figure 3.16).

Discussion

This study presented that the community structure of aquatic organisms in the large scale experiment was significantly affected by the existence of mud snails. It was clear that the addition of mud snails did the change in the community structure of aquatic organisms and their interactions. The existence of mud snails in the paddy field also confirmed that it has effect on the terrestrial community structure and their interactions through rice plant development. It is conceivable that there is bottom up effects in the rice plant ecosystem. In addition, the most abundant group in functional feeding groups in aquatic organisms was the collector including Ostracoda, Copepoda and Oligochaetes. We know that Ostracoda and Copepoda as zooplanktons played an important role to influence both food chain and nutrient cycling in an aquatic ecosystem (Alan et al. 1999; Chittapun et al. 2009). Interestingly, Oligochaetes are one of the important groups and the abundance in benthic fauna of paddy field. These results agree with the observation of Ito et al (2011) showing that Oligochaeta (tubificid worm) population density increased in the paddy fields organically managed.

The present study confirmed that the abundance of aquatic predators species in paddy fields such as diving beetle, water scavenger beetle, leech and other species increased in paddy fields with snail treatment however the patterns were not similar during three years. The interactions between multiple predators can result in facilitation, interference, or neutral relationship (Crowder et al. 1997; Schmitz 2007). Moreover, predators can have different foraging modes that may alter their interactions and impact on food webs (Carey and Wahl 2010), and indirect effects on other components (Schmitz and Suttle 2001; Schmitz 2007). It seems that predators have a strong influence on ecological communities by controlling the abundance and dynamics of species in lower trophic levels (Dobson et al. 2006; Thebault & Loreau 2006). Thus, predation is a key which factor promotes the dynamics of the food web (Carey and Wahl 2010). However, our study provides the evidence that almost all predators tend to be higher in paddy fields with snails and it showed that their abundance was affected by snail addition. This suggests that snail addition increased nutrient supply and enhanced primary productivity, which can directly increase prey abundance, richness and diversity (De Alckmin Marques, Price & Cobb 2000). The prey diversity might influence on abundances of predators which would strongly influence each other.

In this study, the most abundant of functional feeding group in aquatic organisms was collectors including Ostracoda, Copepoda and Oligochaetes. These organisms feed on fine particulate organic matter (FPOM), and also feed on suspension particles of FPOM in the bottom sediments (Merritt et al., 2005). Previous studies confirmed that as zooplankton which they plays an important role to influence both food chain and nutrient cycling (Alan et al. 1999; Chittapun et al. 2009) and Oligochaetes due to organic matter decomposition and nutrient translocation, in aquatic ecosystem (Turner and Ferrante 1979; Vineetha et al. 2015). Based on current information, the abundance of collectors had positively affected by the mud snails. Moreover, several species of collectors such as Ostracoda, Copepoda, Oligochaetes and other insects are also essential prey for aquatic predators.

Scrapers in particular represent a unique group because they feed primarily on algae and associated material (Smith 2016). The previous results demonstrated that benthic invertebrate scrapers were affected on abundance of algae in aquatic ecosystems (Gregory, 1983; Lamberti and Moore, 1984; Wallace, 1996). In this result, the most abundant of scraper was Cladoceran community. It was suggested that the abundance of

Cladocera had positively correlated with mud snail treatment but not with other scrapers. There would be interspecific and intraspecific interactions among scrapers due to algae as food resources. As Cladocera can consume other food resource such as phytoplankton, bacteria or dead plant material (Agasild and Nõges 2005), competition with mud snails might be weak.

Simpson et al. (1994) confirmed that snails play an important role as an indicator of soil fertility in paddy fields. It was suggested that the mud snail addition increased rice plant performance through food web effects and via excretory processes which can release nutrients such as nitrogen and phosphorus in the aquatic ecosystems. The present study showed that plant performance higher than the paddy field without mud snails. The mud snail addition could increase the collectors which were detritus decomposers. Moreover, several studies have reported that activity of collectors affect physical, chemical, and microbiological properties of soil; nutritional status of floodwater and easily uptake of N by rice plants (Grant and Seegers 1985). Hence, the present study suggests that there is bottom up effect in the rice plant ecosystem resulting from direct and indirectly effect of mud snail on rice plant performance, aquatic and terrestrial organisms.

The present study showed that the abundance of terrestrial organisms was higher in the paddy fields with mud snails, although there was no significant difference between two treatments in 2014. In addition, the most abundant group of the feeding guild in terrestrial organisms was other insects. Others or neutral insects in this result are a category that consists of organisms which do not harm to rice plant either directly or indirectly. The previous research demonstrated that plant attributes can affect herbivores, natural enemies, and their interactions are mediated by primary plant attributes (i.e. nutritional quality and physical structure) and other characteristics (Agrawal 2000). In general, this studies showed that snail addition can directly or indirectly affect herbivores and natural enemies. In this context, herbivore-induced changes in terrestrial plants can generate bottom–up trophic cascades from plants to higher trophic levels and can thus influence biodiversity within a community (Ohgushi 2005). Recently, several authors have argued that arthropod communities on plants are structurally organized by plant-mediated indirect effects (Martinsen et al. 2000; Agrawal 2005; Denno & Kaplan 2007; Ohgushi, Craig & Price 2007). Hence, it was clear that snails` activity did change the community structure of terrestrial organisms including herbivores, natural enemies, other/neutral insects and their interactions by altering both rice plant performance and species composition of aquatic organisms

This study confirmed that the addition of mud snails might play an important role in the paddy field ecosystem, in which they can provide suitable habitat for aquatic organisms and also influence the abundance of organisms in terrestrial ecosystems through bottom-up effect.

Table 3.1 List of aquatic organisms found in the field experiment. The (√) indicates that the aquatic organisms was found in the paddy field with and without mud snail

Taxa	Common name	Functional Feeding Group	Mud snails	No Mud Snails
Oligochaetes				
Tubifex tubifex	Sludge worm	C	√	√
Hirudinea				
Glossiphonidae	Leech	P	√	√
Richardsonianidae	Leech	P	√	√
Mollusca				
Sphaeriidae				
Pisidium sp	Pea clams	Sc	√	√
Lymnaeidae				
Radix auricularia japonica	Monoaragai	Sc	√	√
Physidae				
Physa acuta	Bladder snails	Sc	√	√
Coleoptera				
Hydrophilidae				
Berosus punctipennis	Water scavenger beetles	P	√	√
Dytiscidae				
Platambus sp	Predaceous diving beetles	P	√	√
Rhantus suturalis	Predaceous diving beetles	P	√	√
Haliplidae	Crawling water beetles	Sc	√	√
Diptera				
Chironomidae				
Chironomus sp	Chironomid larvae	C	√	√
Ceratopogonidae	Punkies	P	√	√
Ephemeroptera				
Baetidae				
Cleon dipterum	Mayfly larvae	C	√	√
Siphlonuridae				
Siphlonurus sanukensis	Mayfly larvae	C	√	√
Hemiptera				
Gerridae	Water striders	P	√	√
Notonectidae	Backwimmer	P	√	√
Corixidae	Water boatmen	P	√	√
Crustacea				
Ostracoda				
Cypridopsis vidua	Seed shrimps	C	√	√
Fabaeformiscandona myllaina	Seed shrimps		√	√
Copepoda				
Cyclopidae				
Mesocyclops sp	Copepods	C	√	√
Cladocera				
Daphnia sp	Water flea	Sc	√	√
Moina sp	Water flea	Sc	√	√
Loach		P	√	√
Amphibi				
Rana japonica	Tadpole	C	√	√

Abbreviation : P (Predator), C (Collectors including collectors gatherers and filtering collectors, Sc (Scrapers),

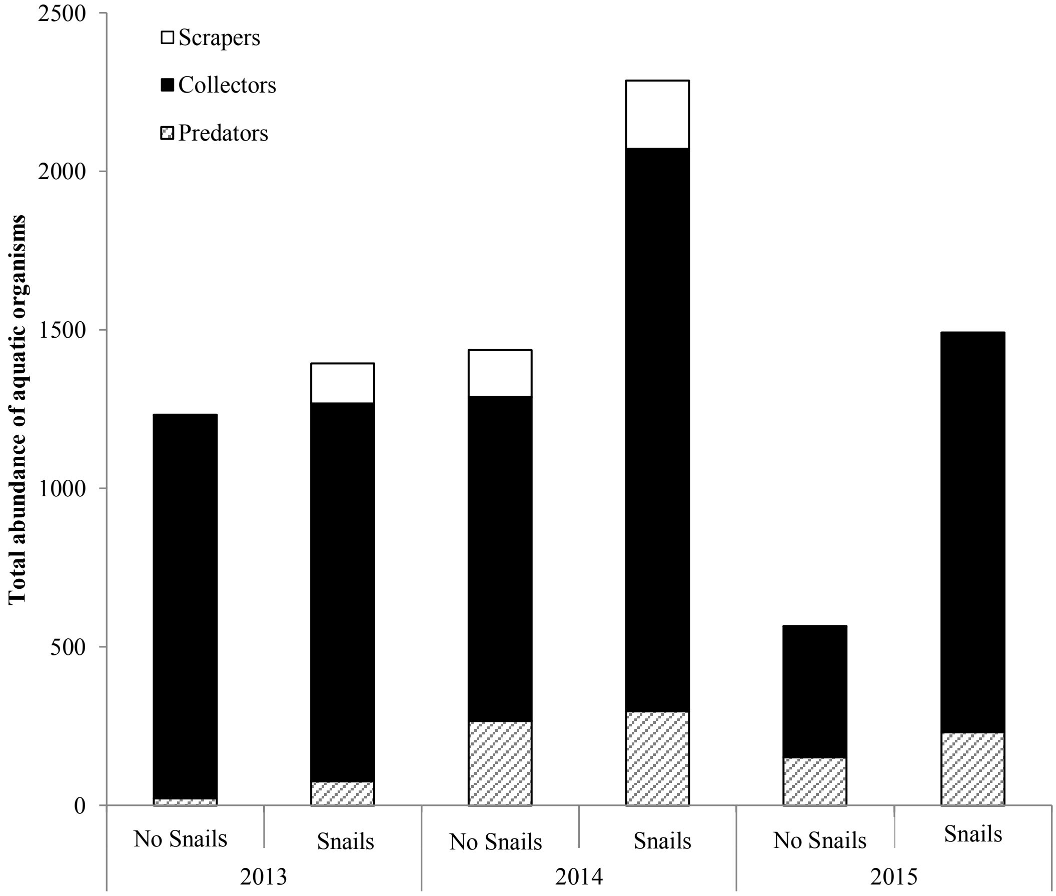


Figure 3.1. Total abundances of predators, collectors, and scrapers in the field with and without mud snails in 2013-2015

a. **2013**

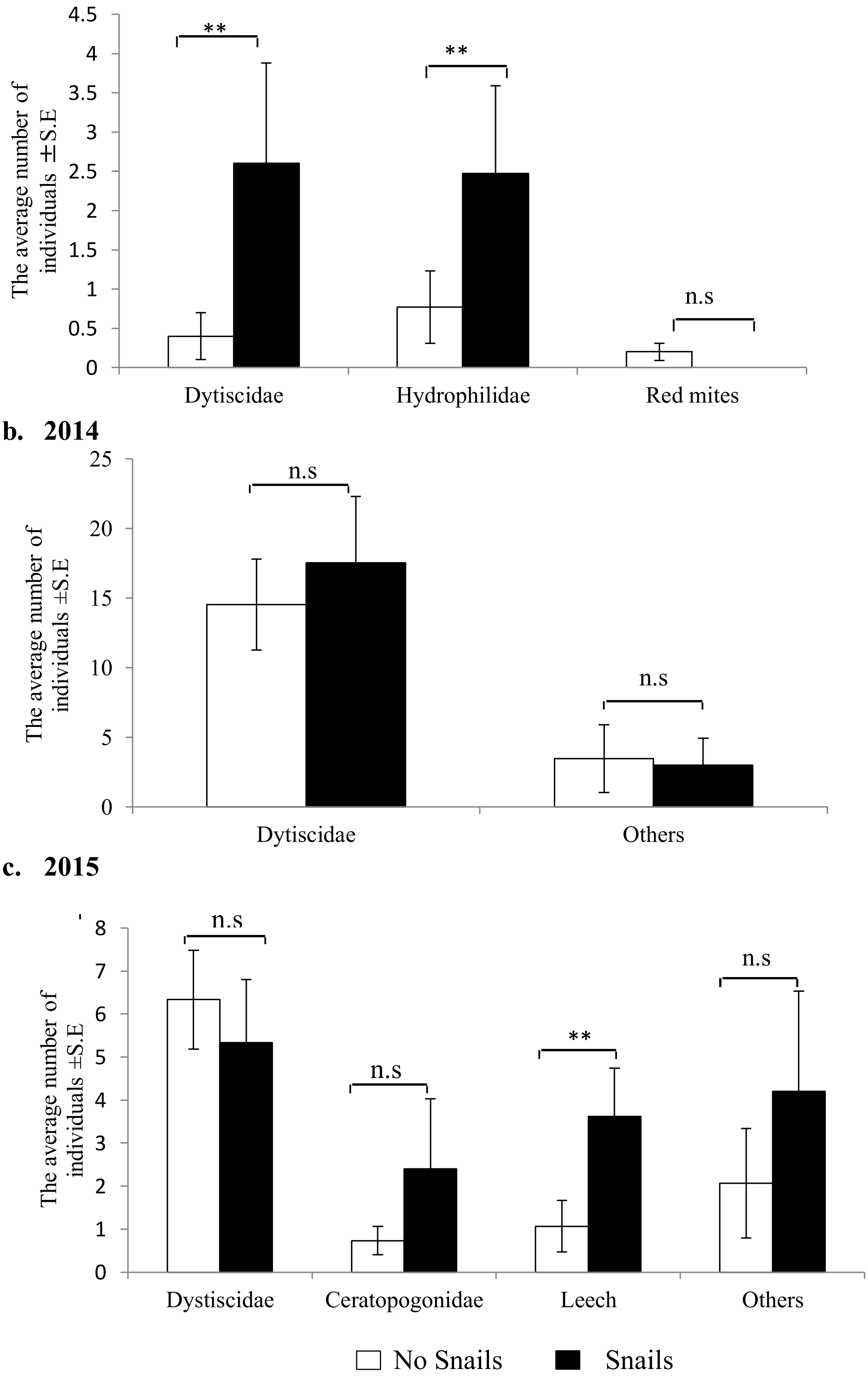
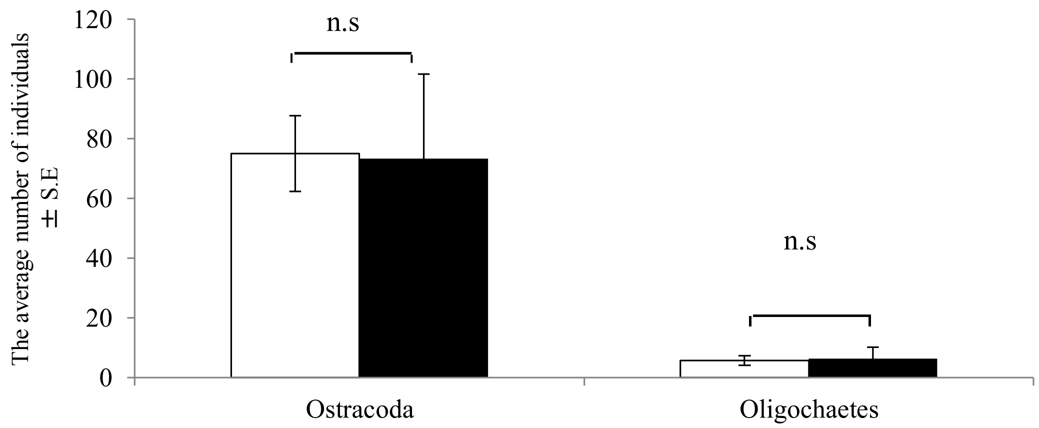
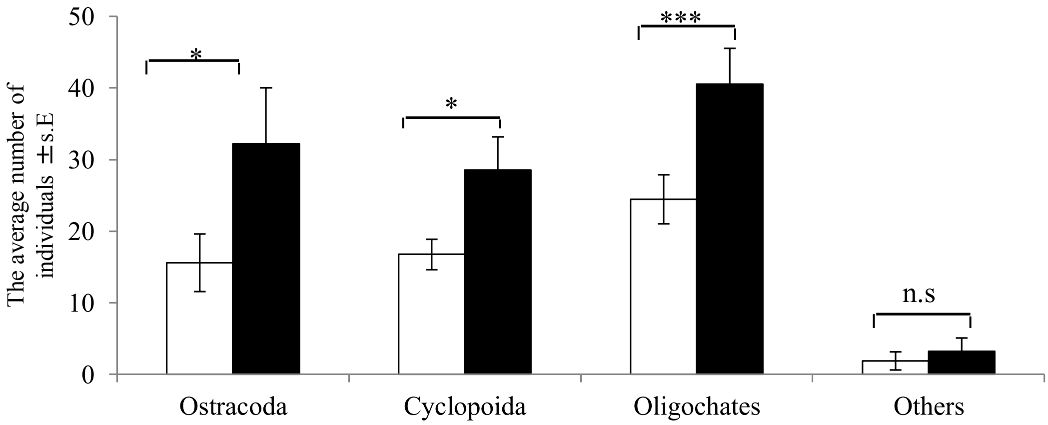


Figure 3.2. The average number (±S.E) of predators in paddy fields with and without snails during 2013-2015. Data were analyzed using the likelihood ratio (LR) test in generalized linier models (GLMs) with poisson and quasipoisson error distribution. (**p<0.01, ns: not significant)

a. 2013



b. 2014



c. 2015

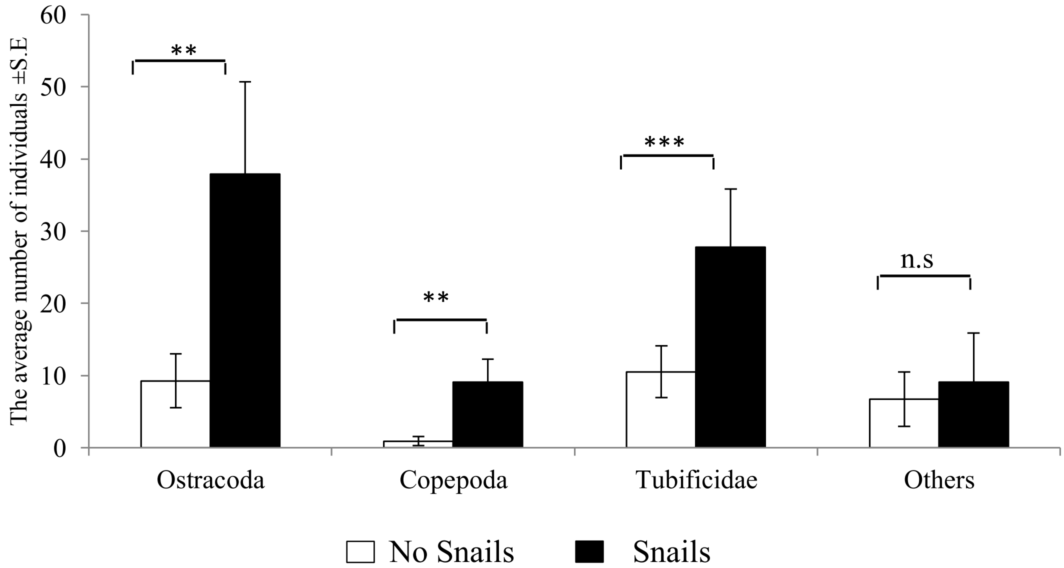
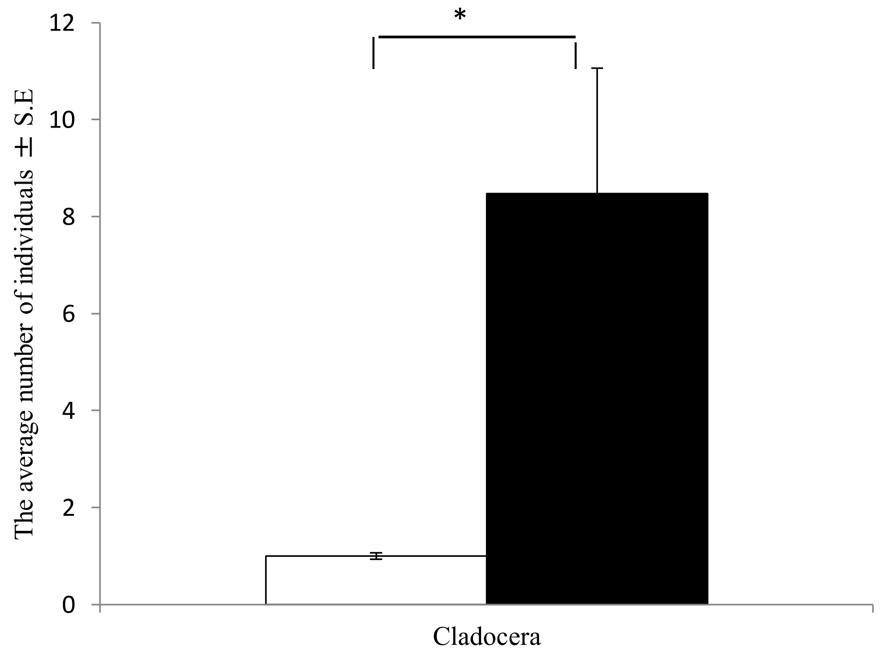


Figure 3.3. The average number (±S.E) of Collectors in paddy fields with and without snail during 2013-2015. Data were analyzed using the likelihood ratio (LR) test in generalized linier models (GLMs) with poisson and quasipoisson error distribution. (****p*<0.001,***p*<0.01,**p*<0.05, ns : not significant)

a. 2013



b. 2014

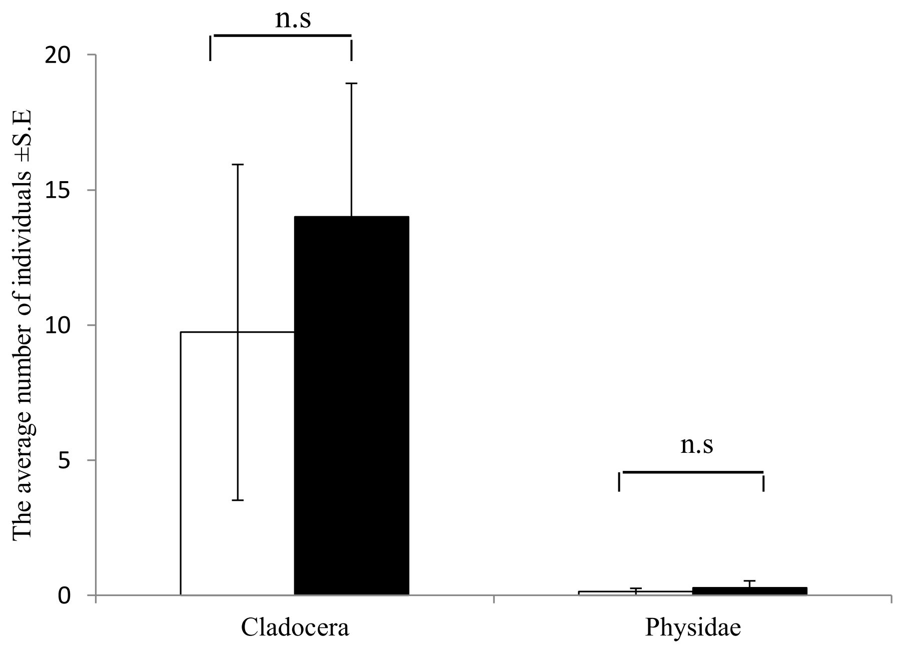
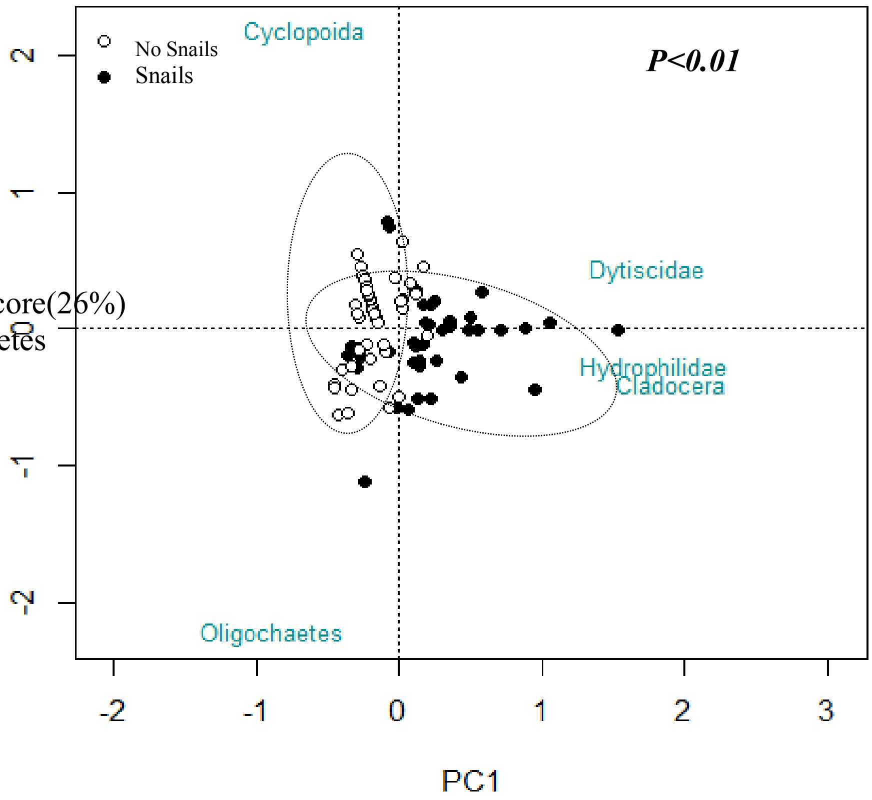


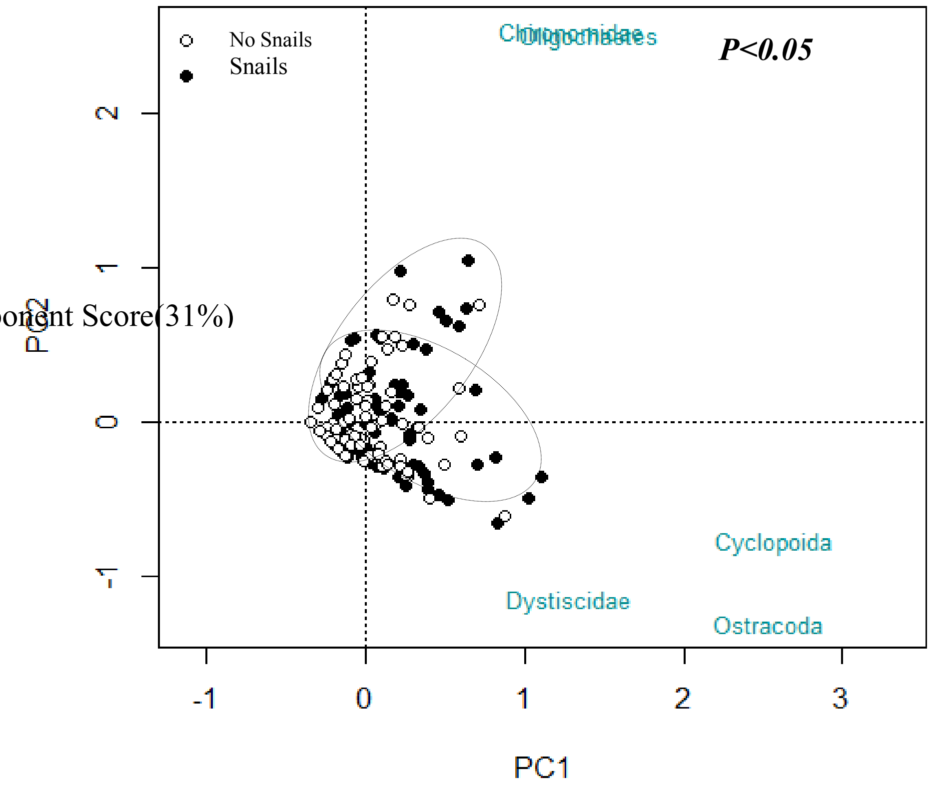
Figure 3.4. The average number (±S.E) of Scrapers in paddy fields with and without snail during 2013-2014. Data were analyzed using the likelihood ratio (LR) test in generalized linier models (GLMs) with poisson and quasipoisson error distribution. (*p<0.05, ns : not significant)

2nd Principal Component Score(26%)
Cyclopoida, Oligochaetes



Cyclopoida, Oligochaetes, Cladocera
1st Principal Component Store (26%)

Fig. 3.5 Principal component analysis (PCA) of the community structure of aquatic organisms in paddy fields with and without snails during in 2013. Figures with % indicate the percentage contributed by the 1st principal component analysis or 2nd one



Ostracoda, Copepoda,Tubificidae
1st Principal Component Store (38%)

Fig. 3.6 Principal component analysis (PCA) of the community structure of aquatic organisms in paddy fields with and without snails in 2014. Figures with % indicate the percentage contributed by the 1st principal component analysis or 2nd one

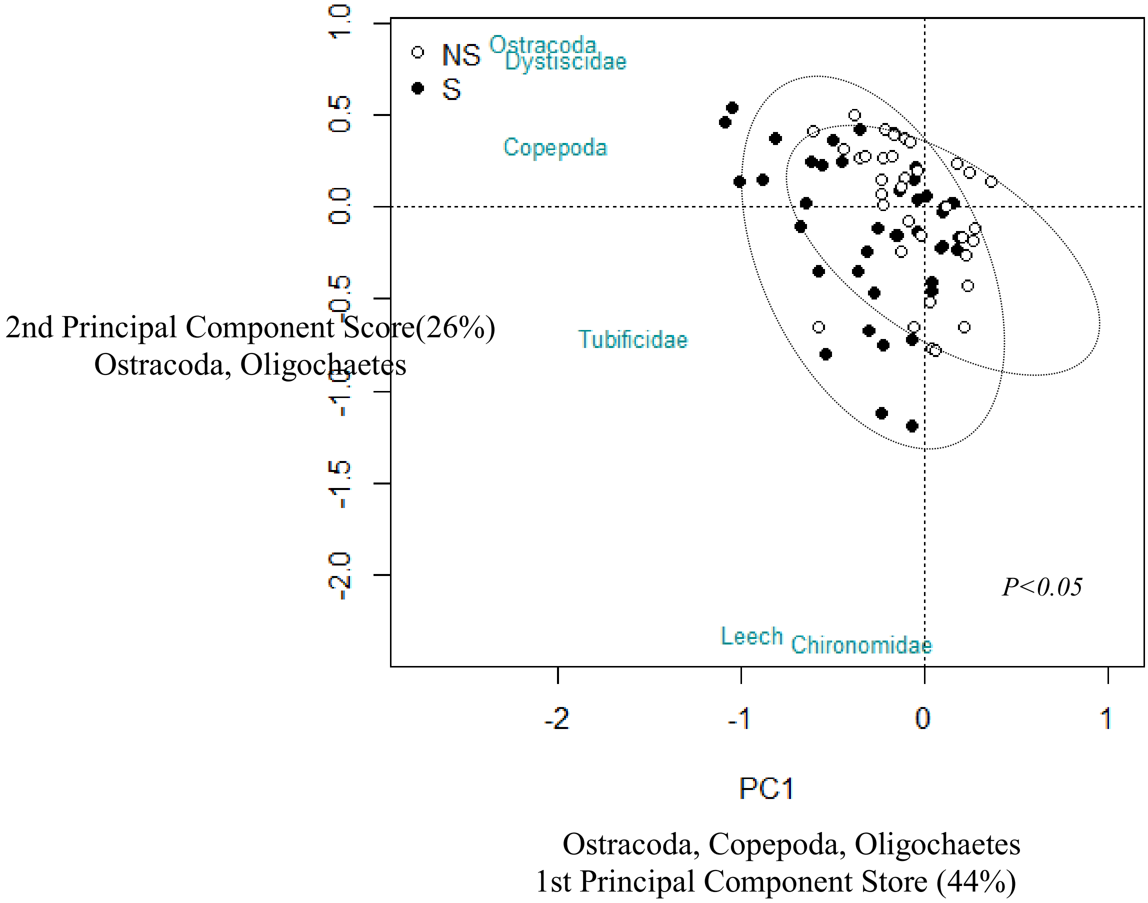


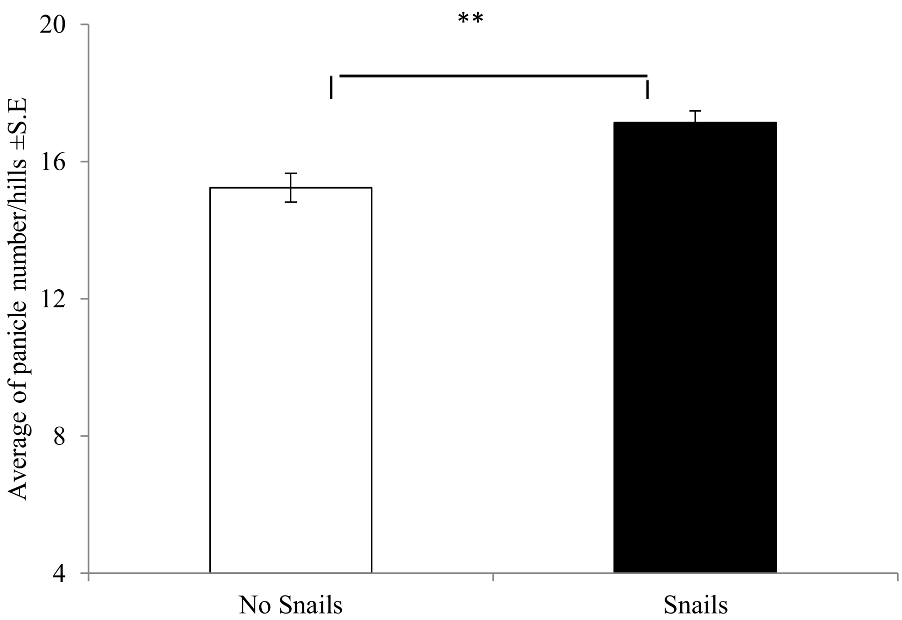
Fig 3.7. Principal component analysis (PCA) of the community structure of aquatic organisms in paddy fields with and without snails in 2015. Figures with % indicate the percentage contributed by the 1st principal component analysis or 2nd one.

Table 3.2 Plant height, tiller number, rice plant biomass/hill, brown rice yields in the plots with and without snails in 2013-2015

Years	Plant height (cm)		Tiller number per hill		Plant Biomass (dry weight) (g)		Brown Rice Yields (g)	
	No Snails	Snail	No Snails	Snails	No Snails	Snails	No Snails	Snails
2013	88.6±0.56a	86.0±0.43b	17.54±0.56a	21.46±0.43b	681.50±36.24a	715.50±27.35a	304.68±17.72a	323.43±16.30a
2014	90.0±0.54a	91.3±0.46a	15.73±0.42a	17.90±0.35b	870.79±49.11a	1018.00±37.17b	336.92±11.57a	394.45±22.14b
2015	101.2±1.53a	101.7±0.76a	18.4±1.38a	23.10±1.02b	736.40±32.24a	833.33±36.54b	278.71±8.90a	324.48±16.76b

Values (mean ± SE) within each column followed by different letters indicate significantly different between the treatments by Welch`s t test (p<0.05). The data of plant biomass was obtained from samplings on September and October in 2013-2015, respectively

(a) 2014



(b) 2015

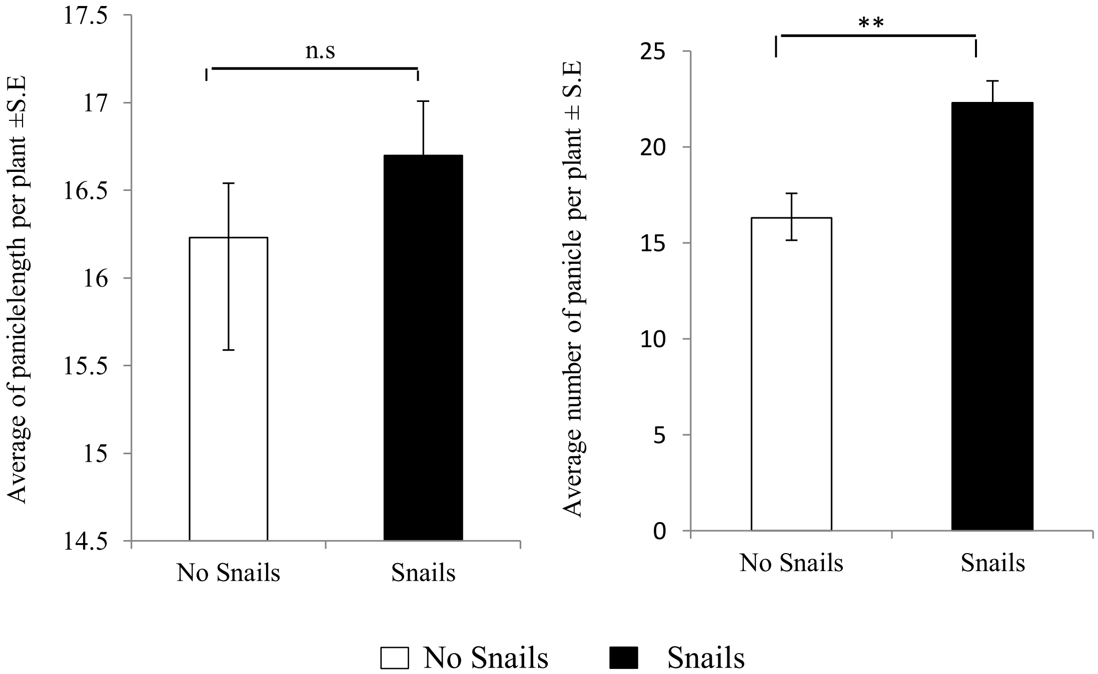


Fig 3-8 Panicle number and panicle length of rice plant by with and without mud snails in 2014-2015. Data were analyzed using Welch`s t test (p<0.05) (**p<0.01, ns : not significant)

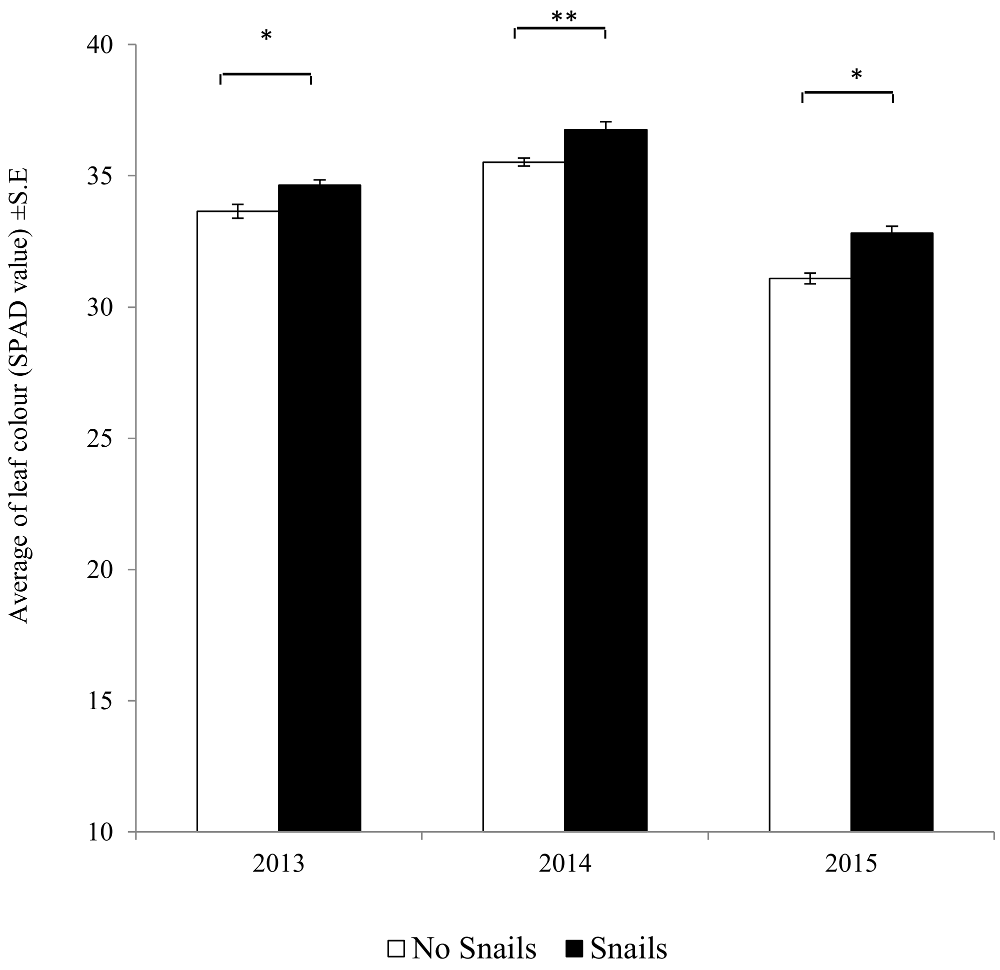


Figure 3-9. Leaf colour (SPAD value) of flag leaves by with and without mud snails in 2013-2015. Data were analyzed using Welch`s t test (**p*<0.05, ***p*<0.01, ns : not significant)

Table 3.3. List of terrestrial organisms found in the field experiment. The (√) indicates that the terrestrial organisms was found in the paddy field with and without mud snail

Taxa	Feeding guilds	Mud snails	No Mud Snails
Hemiptera			
Delphacidae	Herbivore	√	√
Nabidae	Herbivore		√
Coreidae	Herbivore		√
Alydidae	Herbivore		√
Meloidae	Predator	√	
Miridae	Herbivore	√	√
Pentatomiidae	Herbivore	√	√
Homoptera			
Coccidae	Herbivore	√	√
Thripidae	Herbivore		√
Aphididae	Herbivore	√	√
Diptera		√	√
Pomaceae	Herbivore	√	√
Ceratopogonidae	Other insect	√	√
Chironomidae	Other insect	√	√
Tipulidae	Other insect	√	√
Mymariidae	Natural enemies	√	√
Phoridae	Natural enemies	√	√
Syrphidae	Natural enemies	√	√
Dolicopodidae	Natural enemies	√	√
Heleomyzidae	Other insect	√	
Pipunculidae	Other insect	√	
Lauxaniidae	Other insect		
Micropezidae	Other insect	√	
Simuliidae	Other insect		
Chamamyiidae	Other insect	√	
Milichidae	Other insect	√	
Mycetophilidae	Other insect	√	√
Chloropidae	Other insect	√	√
Agromyzidae	Natural enemies	√	√
Shiziomyzidae	Other insect	√	√
Trixosalididae	Other insect	√	√
Sciaridae	Other insect	√	
Scatopsida	Other insect	√	√
Cecidomyiidae	Other insect	√	√
Drosophilidae	Other insect	√	√
Acroceriidae	Natural enemies	√	√
Orthoptera		√	√
Acrididae	Herbivores	√	√
Tettigonidae	Natural enemies	√	
Neuroptera			
Chrysopidae	Natural enemies	√	√
Hymenoptera			
Torymiidae	Natural enemies	√	√
Braconidae	Natural enemies	√	√
Chalcidoidae	Natural enemies	√	√
Eulopidae	Natural enemies	√	√
Eupelmidae	Natural enemies	√	√
Scelionidae	Natural enemies	√	
Proctrupidae	Natural enemies	√	
Tiphidae	Natural enemies	√	
Trichogrammatidae	Natural enemies	√	√
Platygasteridae	Natural enemies	√	√
Cynipidae	Herbivore	√	√
Pteromaliidae	Natural enemies		√
Plecoptera			
Capniidae	Other insect	√	
Perlidae	Other insect	√	
Coleoptera			
Curculionidae	Herbivore	√	√
Coccinelidae	Predator	√	√
Spider			
Tetragnathidae	Predator	√	√
Thomisidae	Predator	√	√
Lynipidae	Predator	√	√
Spider mites	Predator	√	√

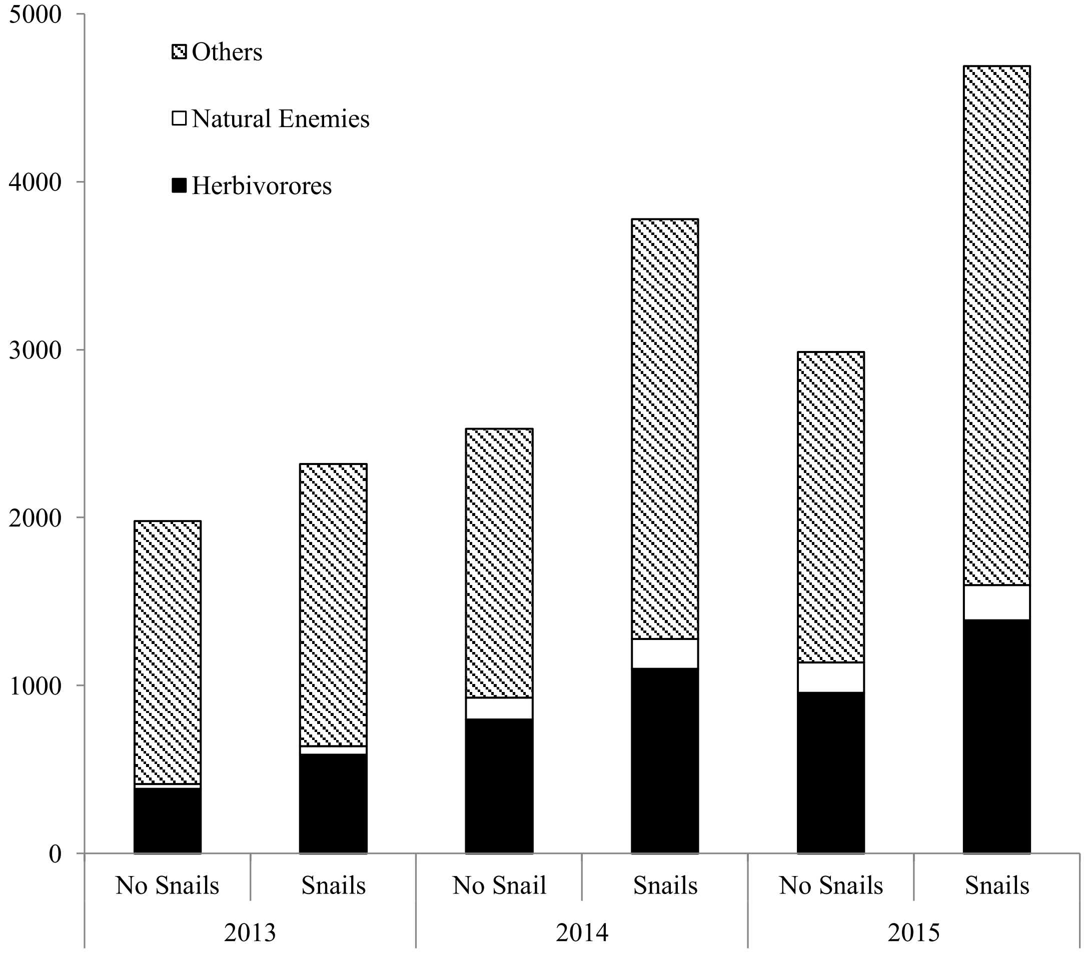


Figure 3.10. Total abundance of herbivores, natural enemies, and other insects in the fieldsby with and without mud snail in 2013-2015

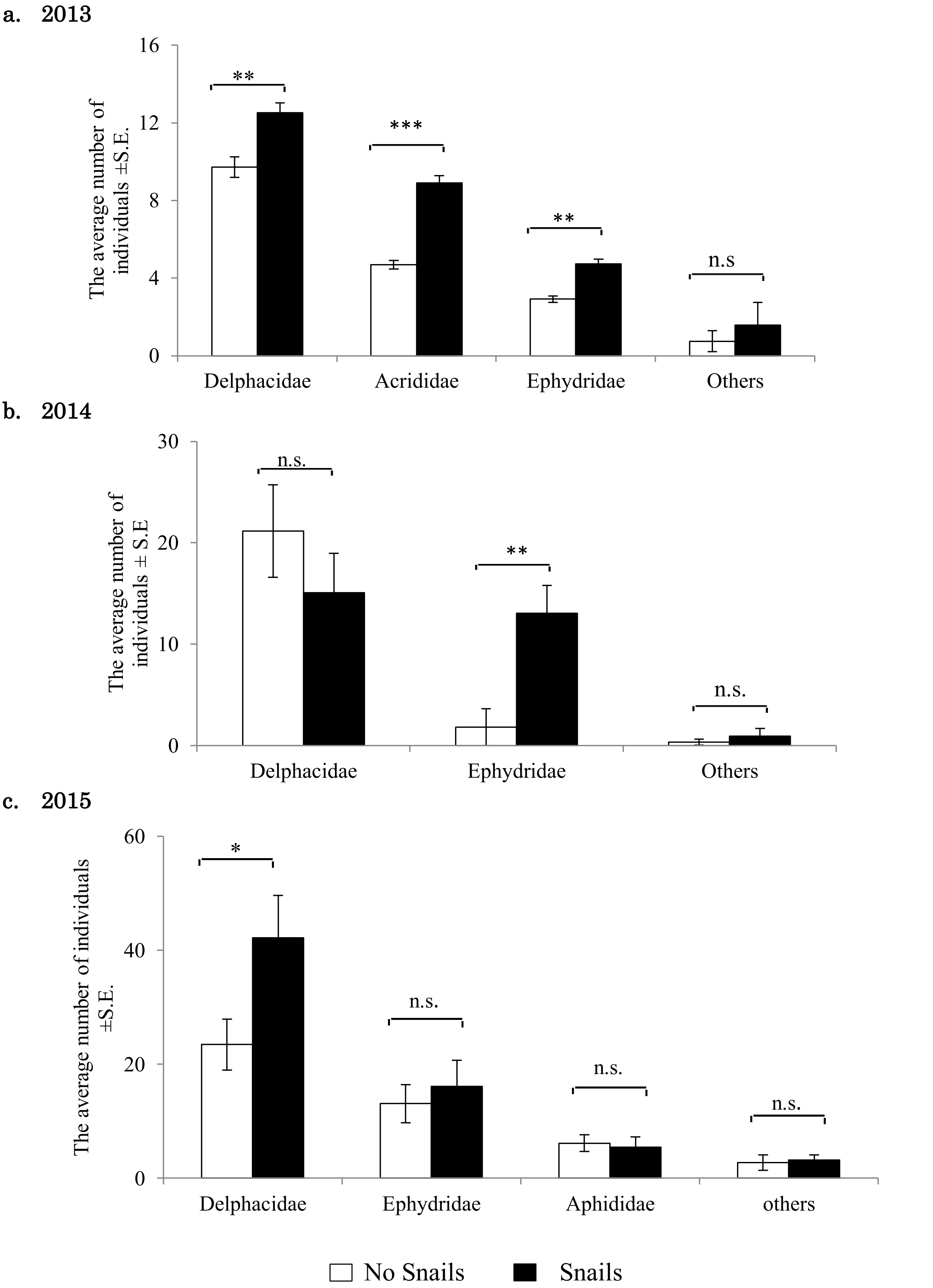


Fig 3.11 The average number (±S.E) of herbivores in paddy fields with and without snails during 2013-2015. Data were analyzed using the likelihood ratio (LR) test in generalized linier models (GLMs) with poisson and quasipoisson error distribution. (***p<0.001,**p<0.01,*p<0.05, ns: not significant)

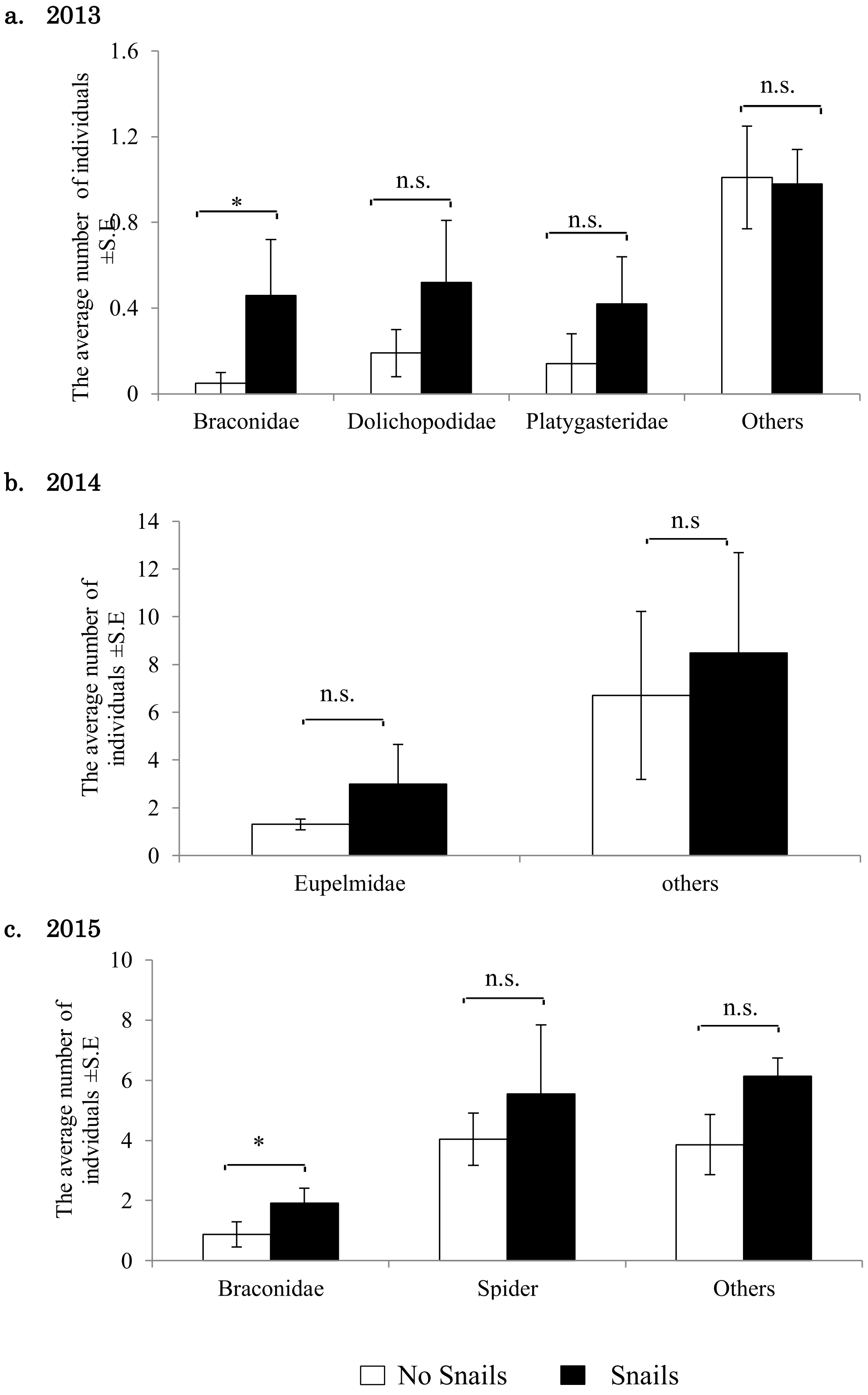


Fig. 3.12 The average number (±S.E) of natural enemies in paddy fields with and without snails during 2013-2015. Data were analyzed using the likelihood ratio (LR) test in generalized linier models (GLMs) with poisson and quasipoisson error distribution. (**p*<0.05, ns : not significant)

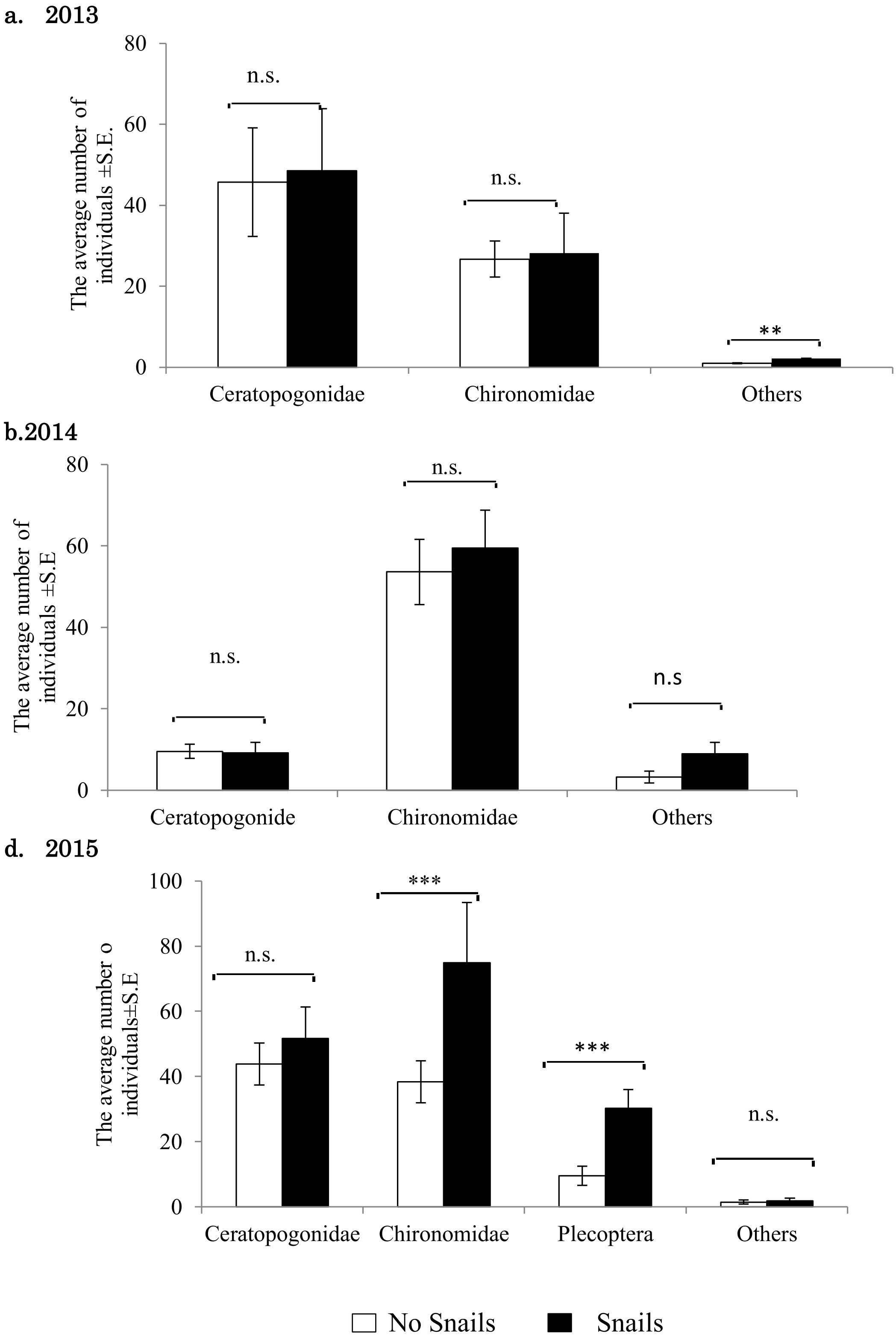
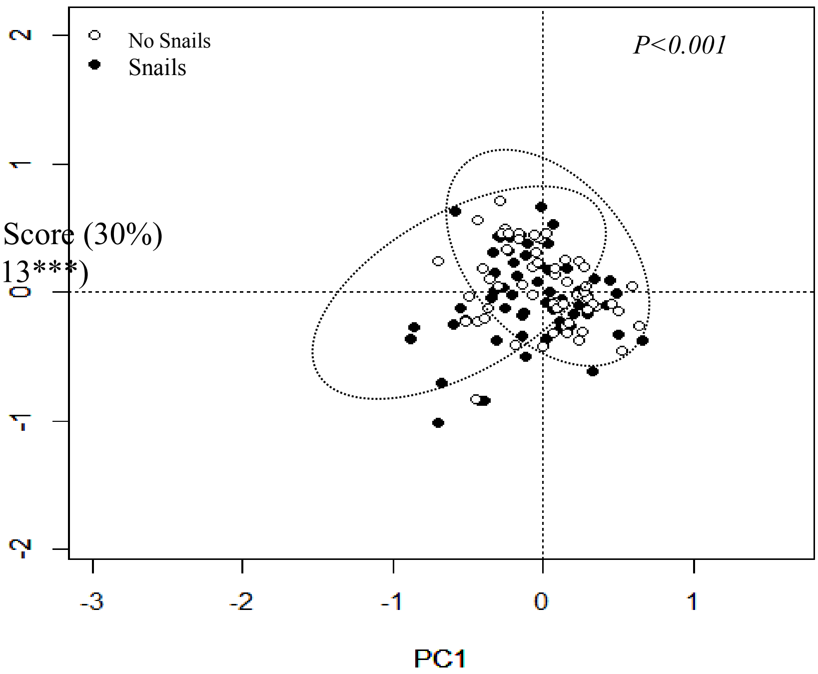


Figure 3.13. The average number (±S.E) of neutral inescts in paddy fields with and without snails during 2013-2015. Data were analyzed using the likelihood ratio (LR) test in generalized linier models (GLMs) with poisson and quasipoisson error distribution. (****p*<0.001,***p*<0.01 and ns: not significant)

2nd Principal Component Score (30%)
Ceratopogonidae (0.13***)



Ceratopogonidae (0.40***), Chironomidae (0.23***)
Delphacidae (0.78***)
1st Principal Component Score (42%)

Fig. 3.14. Principal component analysis (PCA) of the community structure of terrestrial arthropods in paddy fields with and without snail during 2014. Figures with % indicate the percentage contributed by the 1st principal component analysis or 2nd one.

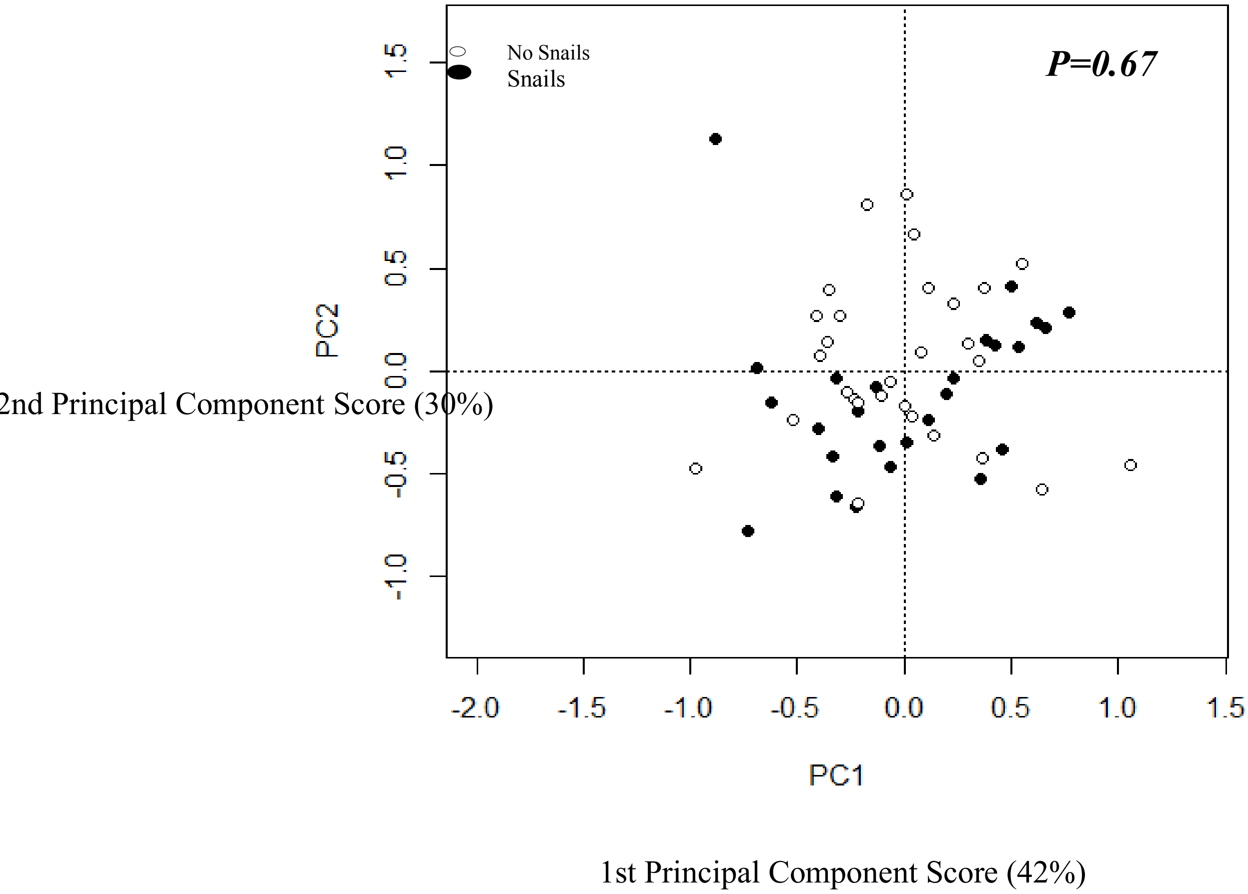
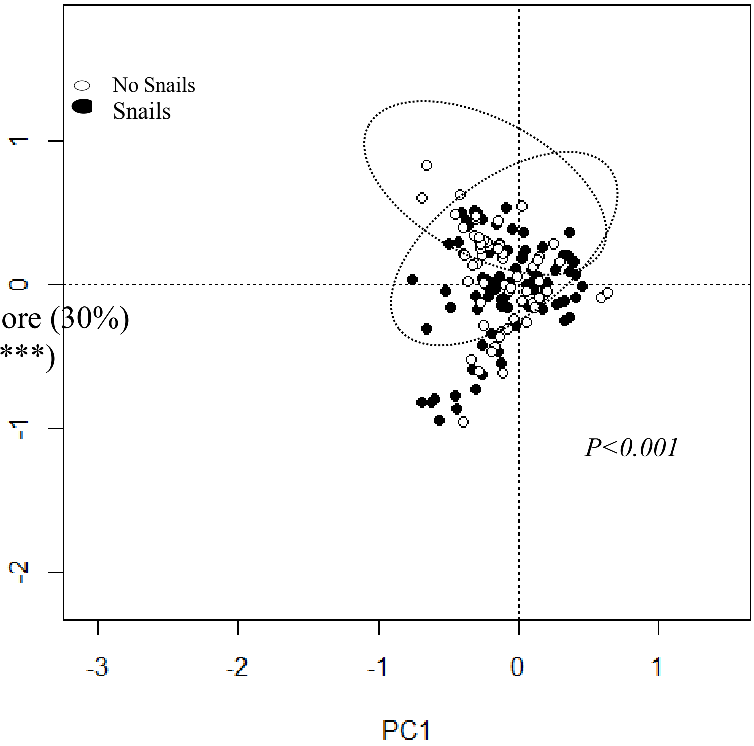


Fig. 3.15. Principal component analysis (PCA) of the community structure of terrestrial arthropods in paddy fields with and without snails in 2014. Figures with % indicate the percentage contributed by the 1st principal component analysis or 2nd one.



2nd Principal Component Score (30%)
Ceratopogonidae (0.13***)

Ceratopogonidae (0.40***), Chironomidae (0.23***)
Delphacidae (0.78***)
1st Principal Component Score (42%)

Fig. 3.16 Principal component analysis (PCA) of the community structure of terrestrial arthropods in paddy fields with and without snail in 2015 Figures with % indicate the percentage contributed by the 1st principal component analysis or 2nd one.

General Discussion

The existence of mud snails in the paddy field also confirmed that it has an effect on the terrestrial community structure and their interactions through rice plant development. It is conceivable that there is bottom-up effect in the rice plant ecosystem including aquatic organisms, rice plant, and terrestrial organisms. This study confirmed that the addition of mud snails might play an important role in the paddy field ecosystem, in which they can provide suitable habitat for aquatic organisms and also influence the abundance of organisms in terrestrial ecosystems through bottom-up effect. In this study, we were used multiple paddy fields to understand more clearly the effects of mud snails in the rice plant ecosystem including community structure in the aquatic and terrestrial ecosystem and their interaction through increasing the rice plant performance. In the previous studies, Leroux and Loreau (2008) also have shown that aquatic and terrestrial food webs can strongly influence each other. For instance, aquatic ecosystems can influence terrestrial ecosystems in at least two different ways: via allochthonous subsidies (Marczak, Thompson & Richardson 2007). Cross-ecosystem organisms are those whose life cycles involve multiple ecosystems.

We understand that rice fields have unique characteristics that make them has a rich biodiversity. However, rice fields constitute man-made ecosystems that dynamic and rapidly changing as consequences of the agricultural development. In fact, conventional agriculture farming, it can lead to a higher yield, due to its extensive use of chemical inputs. However, the chemical inputs would cause several ecological problems (Pimentel et al. 1992). In this study, it was found that developing new concept methods for improving the rice production without using chemical inputs.The new method to minimize the negative effects of chemicals input is using mud snails to release nutrients could improve the rice plant performance. In addition, the effect of mud snails was positively associated with brown rice yields. It was showed that the effect of mud snails was increased gradually on brown rice yield in small and large experiments. Regarding the new concept of environmentally friendly farming, further studies using mud snail as key species and some organisms is needed to understand the more clearly interaction among organisms in multiple ecosystems through direct and indirect interaction at different trophic levels.

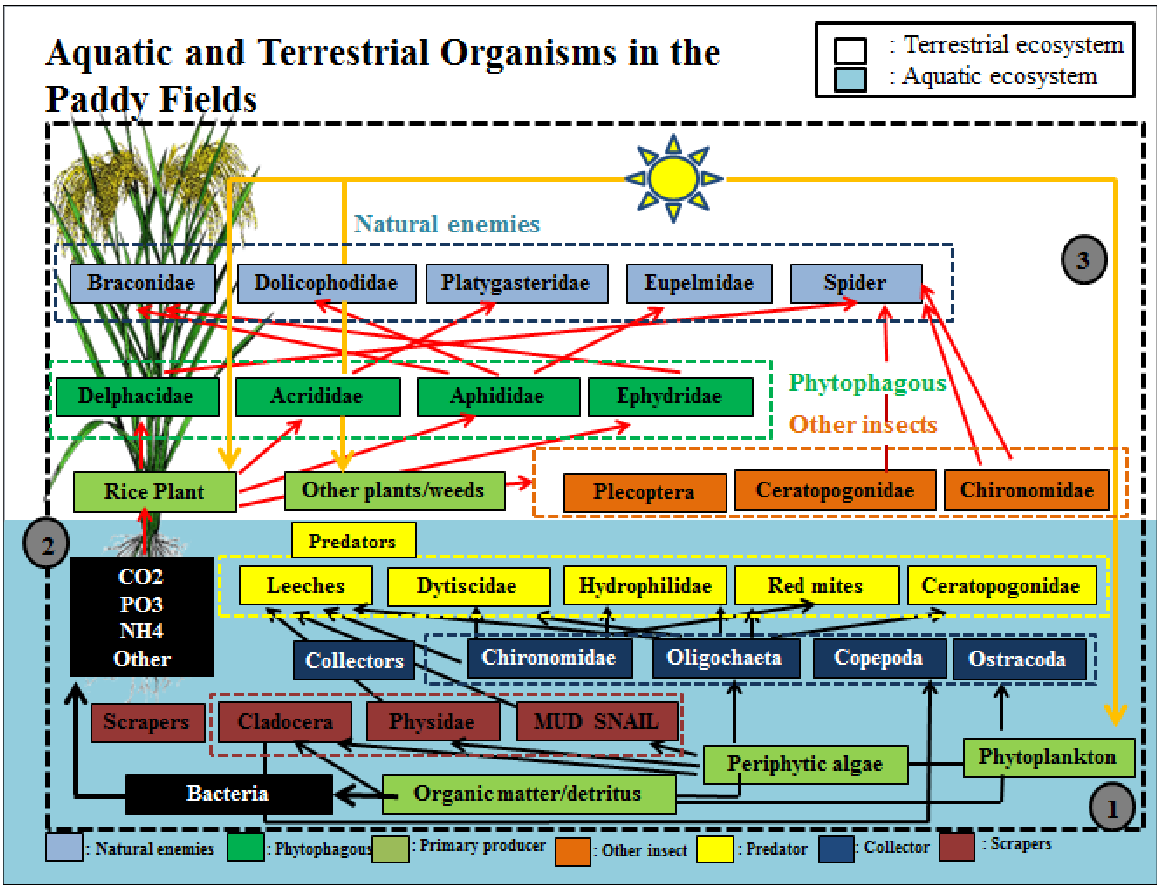


Fig.4.1 Aquatic and terrestrial organisms and their interacton in the paddy fields

As illustrated by a schematic figure, food web theory is based on organisms in the system can be categorized into the trophic level and that organisms at a specific trophic level feed on the trophic level below and fed upon by organisms in the high trophic level (Bronmark and Hansson 2010). In this figure, it will describe the rice ecosystem food web in our experiment. In general, process #1: energy is received by aquatic plant and also stored as organic matter in the soil and brought into the system by micro-organisms and detritivorous insects. The energy flow begins with bacteria being eaten by cladocerans and Ostracoda as zooplankton (Work 2003). Moreover, Funke (2009) noted that bacteria occupy an important role in the aquatic ecosystem since they are major organisms in the decomposition of dead material, and thereby in the recycling of nutrients and carbon. Regarding aquatic organisms that consumed algae and associated materials are referred scrapers such as a freshwater snail. The main food sources of fresh water snails are periphytic algae and detritus (Pinowska 2002). In a parallel flow, most oligochaetes as collectors feed by ingesting the soft sediments that they are often found at high densities in habitat enriched with organic matter. The one of an important role of oligochaetes to promotes release nitrate, phosphate, and potassium in paddy fields. Furthermore, detritivorous insects, such as the larvae of flies (chironomid) feed directly on decaying organic matter, including material floating on the surface of the water and also periphytic algae. Chironomids larvae are also consumed by many invertebrate predators. Based on trophic level, larvae and other organisms (freshwater snail, oligochaetes) also provide a consistent source of food for predators (Coleoptera, fish, and leeches) during rice planting.

In the terrestrial ecosystem, high populations of phytophagous in rice ecosystem result from the fact that good performance of rice plant including plant biomass and other parameters. For natural enemies are directly and not directly dependent on pest populations. Rather, there are three separate avenues for energy flows to natural enemies populations: (1) from organic matter via microorganism cycles and filter -feeding insects, (2) from organic matter via detritivorous insects (neutral insects) and (3) from the rice plant via herbivores. This process is fundamental to aquatic systems and will be found in all rice ecosystems. It was clear that there was mechanisms bottom-up in this experiments.

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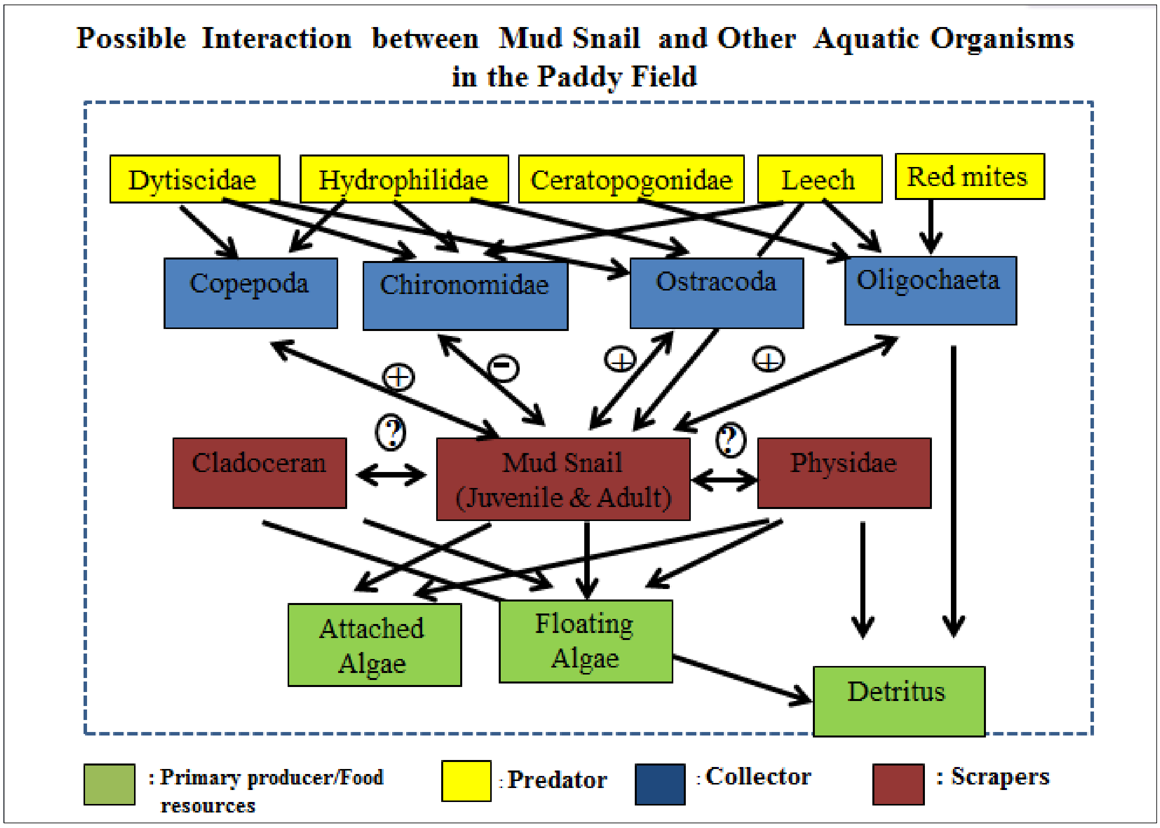

Fig.4.2 Possible interaction between mud snail and other aquatic organisms in the paddy fields

In our study, the most abundant of functional feeding group in aquatic organisms was collectors including Ostracoda, Copepoda, and Oligochaetes. These organisms feed on a fine particulate organic matter (FPOM), and also feeding on suspension particles of FPOM in the bottom sediments (Merritt et al., 2005). Previous studies confirmed that Ostracoda and Copepoda as zooplankton which they play an important role to influence both food chain and nutrient cycling (Alan et al. 1999; Chittapun et al. 2009) and Oligochaetes due to organic matter decomposition and nutrient translocation, in an aquatic ecosystem (Vineetha et al. 2015). Based on current information, the abundance of collectors had positively affected by the mud snails. Moreover, several species of collectors such as Ostracoda, Copepoda, Oligochaetes and other insects are also essential prey for an aquatic predator. Furthermore, scrapers, in particular, represent a unique group because they feed primarily on algae (attached and floating algae) and associated material or detritus (Smith 2016). The previous results demonstrated that benthic invertebrate scrapers both are affected by and have an effect on algae in aquatic ecosystems (Gregory, 1983; Lamberti and Moore, 1984; Wallace, 1996). In this result, the most abundant of scraper was Cladoceran community. It was indicated that the abundance of Cladocera had positively correlated with mud snail treatment but not with other scrapers. There was interspecific and intraspecific interaction among scrapers due to algae as food resources. It was suggested that Cladocera can consume other food resources such as phytoplankton, bacteria or dead plant material (Agasild and Nõges 2005) and no competition with mud snail. In addition, there were not dominant species in predator because the available and diversity of prey.

Summary

The purpose of this study was to understand the effects of the mud snails on aquatic invertebrates and terrestrial arthropods through the snails' effects on rice plant development in the paddy fields. Thus we compared the abundance of aquatic organisms and terrestrial arthropods, and rice plant performance, in response to a field experiment with and without mud snails in a paddy field. The purpose of this study was to examine (1) how the effects of the mud snails on the community structure of aquatic organisms and (2) how the community of aquatic organisms affected on the terrestrial organisms communities through the rice plant development in the paddy field (Small and large scale experiments). Furthermore, this study also to understand applied aspect from two experiments, how the mud snails influence on rice yields without chemical inputs during rice o farming. Is it possible or not?. We hypothesized that the mud snail would increase aquatic and terrestrial community structure and rice plant development through the year. This study clearly showed that the community structure of aquatic organisms in the rice paddy plots was not significantly altered by the addition of mud snails, but this addition did change the community structure of terrestrial organisms as associated with a positive influence on rice plant growth. Hence the mud snails appeared to have bottom up effects on terrestrial organisms resulting from the effects of the mud snails on rice plant development. This study suggested that mud snails might play an important role in the paddy field ecosystem, in which they influence the abundance of organisms in terrestrial ecosystems through direct and indirect interactions at different trophic levels. Further investigation using multiple paddy fields is needed to understand the ecological processes underlying the effects of mud snails on arthropods, e.g. aquatic nutrient recycling, in paddy fields since present study was carried out as a small-scale experiment.

To clarify how the interaction of aquatic organisms which directly or indirectly influence on the abundance of terrestrial arthropods through rice plant performance in the multiple paddy fields in large scale experiment, the field experiments were conducted in the field by with and without snails. The purpose of this study was to understand (1) how the effects of the mud snails on the community structure of aquatic organisms and their interaction, and (2) how the community of aquatic organisms affected on the terrestrial organisms communities through the rice plant development in the paddy field (field experiment). Thus we compared the abundance of aquatic organisms and terrestrial arthropods, and rice plant performance, in response to a field experiment with and without mud snails in a paddy field. We hypothesized that the addition of mud snail would more clearly increase aquatic and terrestrial community structure and rice plant development through the year in the large scale experiment. The result showed that the community structure of aquatic organisms in the large scale experiment was significantly affected due to the existence of mud snails. It was cleared that the addition of mud snails did the changed the community structure of aquatic organisms and their interactions. The existence of mud snails in the paddy field also confirmed that it has effect on the terrestrial community structure and their interactions through rice plant development. It is conceivable that there is bottom effect in the rice plant ecosystem.

This study confirmed that the addition of mud snails might play an important role in the paddy field ecosystem, in which they can provide suitable habitat for aquatic organisms and also influence the abundance of organisms in terrestrial ecosystems through bottom-up effect. In this study, we were used multiple paddy fields to understand more clearly the effects of mud snails in the rice plant ecosystem including community structure in aquatic and terrestrial ecosystem and their interaction through increasing the rice plant performance.

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