Community structure and seasonal changes in aquatic oligochaetes in an organic paddy field in Japan

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Abstract Field surveys of aquatic oligochaetes were conducted in an organic paddy field once per month in Kamakura, central Japan between June 2010 and May 2011, except during winter. We found eight aquatic oligochaete taxa. The species composition agreed with results from other paddy fields and eutrophic lakes in Japan. The oligochaete community was dominated by the tubificine Limnodrilus hoffmeisteri and the rhyacodriline Branchiura sowerbyi. The tubificine Embolocephalus yanaguchii was also collected. This species has been most frequently reported from springs and their outlet streams. Total population density and biomass were 2800 m$^{-2}$ and 21.2 g wet wt. m$^{-2}$, respectively, lower than those in other similar paddy fields in northern Japan and the Philippines. The population dynamics of B. sowerbyi were examined by size distribution. Growth and reproduction continued until September; however, growth was reduced beginning in October. B. sowerbyi overwintered at various development stages, and newly born individuals were found beginning in April. The management practice for this paddy (organic, winter flooding) seemed to disturb aquatic oligochaeta less than does conventional farming (chemical, winter drainage), therefore the population remained stable throughout the year.

Key words: Aquatic oligochaetes, Branchiura sowerbyi, Organic farming, Paddy field

Introduction

Rice cultivation in Japan has a history of more than 2400 years. In conventional farming, the use of agricultural chemicals to control rice pests significantly increases grain yields. For paddy rice production in Japan, many farmers use herbicides to reduce total labour. By using herbicides, working hours were reduced from 50 hours to 2 hours per year, between the 1950s and 2000 (The Ministry of Agriculture, Forestry and Fisheries of Japan, 1951-2000). However, because chemicals are often nonspecific, they have the potential to affect non-target species (Roger et al., 1994; Simpson and Roger, 1995). The risk assessment for the use of chemicals in Japan has been designed to protect aquatic organisms under the Agricultural Chemicals Regulation Law. Three main taxonomic groups representing different trophic levels are considered including fish, crustaceans (daphnids), and algae. Under the law, a risk assessment is performed by comparing the acute effect concentration of a chemical with predicted environmental concentrations calculated using environmental models for the chemical concerned (The Ministry of the Environment, Government of Japan, 2005). This risk-assessment method has been established for animals inhabiting surface waters. In contrast, the impacts on paddy soil biota, such as aquatic oligochaetes, and the insect fauna of paddy fields outside the paddy water are not considered by this policy. Therefore, such a risk assessment does not necessarily provide adequate protection for all living organisms in paddy fields.

Aquatic oligochaetes are a major component of the invertebrate fauna of flooded paddy fields but they have received little research attention (Simpson et al., 1993a). Aquatic oligochaetes have mainly been studied in lakes (Ohtaka and Kikuchi, 1997; Ohtaka and Nishino, 1999; Raburu et al., 2002). For example, the population density of one species of aquatic oligochaete was 27,600 m$^{-2}$ in eutrophic
Lake Kitaura, central Japan (Ohtaka and Kikchi, 1997). In a previous study performed in Japanese paddy fields, the density of aquatic oligochaetes increased from only a few at the time of crop transplant to 50,000 m^-2 in the middle of July (Kikuchi et al., 1975). These studies indicate that aquatic oligochaete density in paddy fields is comparable to that in eutrophic lakes. Including aquatic oligochaeta, the macroinvertebrate community is an important trophic level in wetland systems, providing food for several wildlife species, such as fishes and waterfowl (Wissinger, 1999). Aquatic oligochaetes in paddies are attracting the attention of researchers as part of a new agricultural method (organic agriculture or agriculture with environmental conservation) that relies on their ecosystem function, and a few farmers are using these methods or showing interest. However, compared to conventional agriculture, the rate of diffusion is only 0.1 per cent (as of the end of 2004 in Japan) (The Ministry of Agriculture, Forestry and Fisheries of Japan, 2004a-c). The role of aquatic oligochaetes is exerted through respiration, burrowing, ingestion, digestion, and excretion (Simpson et al., 1993a). Under laboratory conditions, aquatic oligochaetes promote nutrient mineralisation (Kikuchi and Kurihara, 1977; Roger et al., 1987) and suppress weed germination (Kikuchi et al., 1977). Therefore, although the organisms are of small size, aquatic oligochaetes may play a substantial role in plant growth in paddies (Kikuchi et al., 1977; Kurihara and Kikuchi, 1988). Population density or diversity changes in aquatic oligochaetes will reflect agricultural practices. When flooding paddies without using chemicals, the population density of aquatic oligochaetes was seven times higher than during conventional agriculture (Ito et al., 2011). Despite numerous laboratory tests, few studies have examined oligochaeta population density, geographical distribution, species composition, and population dynamics in paddy fields (Kikuchi and Kurihara, 1977; Simpson et al., 1993a, b; Yokota and Kaneko, 2002).

The objective of this study was to determine the structure of the aquatic oligochaeta community (i.e., species composition, dominant species, richness, abundance, and biomass) and the seasonal changes in a paddy field. Research on aquatic oligochaetes in organically farmed paddy fields will contribute to a better understanding of the effects of modern agricultural practises on paddy biota. These data can be compared with data from previous studies on conventional paddy fields and used to assess the ecological risks of spraying agricultural chemicals.

Materials and Methods

Study area
Field surveys were conducted at an organic paddy field once per month during June 2010 and May 2011, except during the winter (December 2010 to February 2011) in Kamakura central park (56 m above sea level, 139°52′ N, 35°33′ E) of Kanagawa Prefecture, central Japan. Activity and growth of aquatic oligochaeta (observed in this site) decreases under 13°C. (Aston et al., 1982; Nascimento and Alves, 2009). Therefore, we assumed that aquatic oligochaeta could maintain a state of November 2010 and no sampling has been done during the winter season in this study. This paddy has been maintained traditionally for about 650 years. The construction of Kamakura central park started in 1991 and was completed in 1997, and the paddy fields were maintained by farmers during the construction period. Since the park opened in 1997, the paddy has been maintained by a non-profit organisation using traditional farming practises and has also been used for educational purposes. This paddy field has no modern facilities and it has been maintained using traditional Japanese methods (e.g., levees are made of hardened paddy soil). No machines are used except for paddling (Table 1). The paddy is located in a scenic park surrounded by a hilly area, and no private houses are situated within a 200 m radius of the paddy. Water irrigation follows a “plot-to-plot irrigation system” irrigation water is obtained from a hill stream at higher elevation fields; the water travels to lower elevation fields through these upstream paddy fields by cutting a part of the levees surrounding the paddy plots and letting the excess flow into downstream paddy fields. Four paddy fields are situated upstream and their agricultural management methods are the same as those of the study site. Water management is performed in winter-flooded paddy fields, as the paddies are flooded throughout the year. It is unclear whether pesticides were used in the study area before 1997. However, pesticides (Baycid EC and Disyston Gtunule, Bayer Crop Science, Monheim am Rhein, Germany) were used from 1997 to 2003. Neither pesticides nor chemical fertilisers have been used for more than 8 years, including during the monitoring period.

Sampling design
Paddy soil samples were collected from the site throughout the year using a hand-held corer (15 cm long and 6 cm in diameter). The corer was positioned within 10 cm
around rice plants. After dividing the paddy field (136 m²) into 25 sections (3 m² each), 20 were chosen at random, and approximately 500 g of soil were collected. The collected samples were brought back to the laboratory, and divided in half based on weight. One half was used for measuring the abundance and biomass of aquatic oligochaetes, and the other half was used for measuring the properties of paddy soil and for analysing aquatic oligochaete species composition.

Abundance and biomass of aquatic oligochaetes

Aquatic oligochaetes were extracted from each of the substrates using a Baermann apparatus (706.9 cm²) with 40 W electric bulbs for 2 days and then preserved in water. After extraction from the paddy soil, the numbers of individual aquatic oligochaetes were counted in a living state. A 10× magnification digital microscope (VHX1000, Keyence Co., Tokyo, Japan) was used for image acquisition. All images were processed using software (VHX1000, Keyence) to estimate oligochaete biomass from the projected area of the image. The oligochaetes used to determine the projected area were collected by hand-sorting from the study sample, and used to estimate the biomass of oligochaetes in the projected area. Aquatic oligochaetes were raised for 48-h in deionised water to excrete intestinal residue. After 48-h, these aquatic oligochaetes were filmed while alive using a 10× magnification digital microscope. Fresh weight was recorded after filming, while the specimens were still alive. Excess water was removed by placing the specimens on filter paper. The generalised linear model obtained by linear regression from the correlation between the projected area and wet weight using a gamma distribution is presented in Fig. 1.

We measured the dominant species in the study area, which included Limnodrilus spp. and Branchiura sowerbyi Beddard, 1892. Two regression formulas were developed for each dominant species. The biomass of all oligochaetes other than B. sowerbyi was estimated using the formula for Limnodrilus spp. The size class distribution of B. sowerbyi was analysed, because it is possible to identify B. sowerbyi juveniles. B. sowerbyi was weighed by size class (0–10, 10–20, 20–30, 30–40, 40–50, 50–60, and 60–70 mg), which revealed a seasonal variation in size.

Analysis of aquatic oligochaete species composition

Species composition was determined by sieving sediment through a 0.25-mm mesh sieve. Sorting was performed by hand using fresh samples when the organisms were mobile, which contributed to quicker and more efficient sorting. After sieving, the specimens were washed with distilled water and filmed using a 10× magnification digital microscope while still alive, before preservation in 10% formalin for sample preparation. Identification was completed for oligochaete slide specimens that had first been dehydrated in a graded series of ethanol and water solutions, cleared in methyl salicylate and then mounted as whole specimens on slides in Canada balsam. Detailed observations of chaetal and genital morphologies were made using an optical microscope (BX50-33 PHD, Olympus Co., Tokyo, Japan). All specimens were identified based on the literature (Brinkhurst and Jamieson, 1971; Ohtaka, 1994; Kathman and Brinkhurst, 1999). We examined all of the aquatic oligochaete samples collected to analyse seasonal species composition. However, the density was very high in November 2010 and May 2011, therefore, we selected
100 specimens randomly from each of 20 samples. The family and subfamily taxonomy of the oligochaetes followed the classification system of Erseus and Gustavsson (2002) and Erseus et al. (2008).

Environmental data

We measured water temperature, conductivity (D-54T, HORIBA Ltd., Tokyo, Japan), pH (D-21, HORIBA) and dissolved oxygen (DO24P, DKK-TOA Co., Tokyo, Japan) of the irrigation inflow, the paddy water, and the drainage outflow water on each sampling date. Water samples were brought back to the laboratory, where nitrate concentrations were measured (DX320, Dionex Co., Sunnyvale, CA, USA) (Table 2). Soil nitrogen and carbon content was measured using a CN recorder (MACRO CORDER JM1000CN, J-Science, Kyoto, Japan) (Table 2). Paddy soil temperature was measured at depths of 0 and 10 cm using a data logger (Thermocron, OnSolution Pty, Ltd., Baulkham Hills, NSQ, Australia) (Fig. 2).

Statistical analysis

Soil nitrogen concentrations and soil carbon concentrations were analysed using Games–Howell tests at p=0.05. Data were analyzed using R 2.12.1 for Microsoft Windows (R Development Core Team, 2010).

Results

Concentration of soil nitrogen and carbon

The soil nitrogen concentration in November 2010 (after harvest) was significantly higher than during July and August 2010 (p<0.05). In March 2011, soil nitrogen was significantly higher than before harvest, in June to October 2010 (p<0.05) (Table 2). Soil carbon concentrations showed a similar pattern to the nitrogen concentrations; levels were significantly higher in November 2010 than in July 2010 and were significantly higher in March 2011 than in June, August and October 2010 (p<0.05) (Table 2).

Biomass estimation

The biomass used for the projected area measurements ranged from 0.29 to 8.06 mg and 1.16 to 58.76 mg for Limnodrilus spp. and B. sowerbyi, respectively. The following formulae were obtained from linear regressions of projected area and wet weight (Fig. 1).

\[ \text{Limnodrilus spp.} \]
\[ \text{Wet wt. (mg)} = -0.124 + 0.392 \times A \quad R^2 = 0.977 \quad (1) \]

\[ \text{B. sowerbyi} \]
\[ \text{Wet wt. (mg)} = -2.79 + 0.699 \times A \quad R^2 = 0.973 \quad (2) \]

where "A" is projected area (mm²)
Table 2. Seasonal changes in water properties of inflow, paddy and outflow water, and soil nitrogen and carbon concentration at Kamakura paddy field. Field surveys were conducted once a month between 2010 and 2011 except winter (December 2010 to February 2011).

<table>
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<th>Parameters</th>
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<td></td>
<td>Jun Jul Aug</td>
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<td></td>
<td>Sep</td>
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<tr>
<td>a) Irrigation inflow</td>
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</tr>
<tr>
<td>Water temperature (°C)</td>
<td>20.6 25.0 26.3</td>
</tr>
<tr>
<td>pH</td>
<td>6.8 6.8 6.7</td>
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<td>DO (mg L⁻¹)</td>
<td>6.0 4.7 4.3</td>
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<tr>
<td>NO₃⁻ (mg L⁻¹)</td>
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</tr>
<tr>
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<td>b) Paddy water</td>
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<tr>
<td>Water temperature (°C)</td>
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<tr>
<td>pH</td>
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<td>NO₃⁻ (mg L⁻¹)</td>
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<td>Conductivity (mS m⁻¹)</td>
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<tr>
<td>c) Drainage outflow</td>
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<tr>
<td>pH</td>
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<td>NO₃⁻ (mg L⁻¹)</td>
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<tr>
<td>Conductivity (mS m⁻¹)</td>
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<tr>
<td>d) Paddy soil (n = 20)</td>
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<td>± SE</td>
<td>±0.01 ±0.01 ±0.01</td>
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<tr>
<td>Carbon (kg m⁻²)</td>
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<td>± SE</td>
<td>±0.07 ±0.04 ±0.06</td>
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Fig. 2. Seasonal changes in soil temperature at the Kamakura paddy field between 2010 and 2011. Data were missing during the period between October 11 and 15.
Table 3. Species composition of oligochaetes at Kamakura paddy fields. Field surveys were conducted once a month between 2010 and 2011 except winter (December 2010 to February 2011). Immature individuals were identified from the setae. Figures in parenthesis are matures.

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<tr>
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<td>16</td>
<td>(13)</td>
<td>2</td>
<td>(2)</td>
<td>5</td>
<td>(4)</td>
<td>10</td>
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<td>(Brinkhurst, 1971)</td>
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<td>(4)</td>
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Fig. 3. Seasonal changes in the population density of aquatic oligochaetes at the Kamakura paddy field. Field surveys were conducted once a month between 2010 and 2011, except in the winter (December 2010 to February 2011). Mean (n=20) and S.E. (bar) values are shown. a) Total Oligochaeta, b) B. sowerbyi.

Species composition

Immature worms were found throughout the year. The dominance of the immature worms in each month calculated it without including a population of "Unknown". The highest proportion of immature worms (90%) occurred in June and October 2010, and the lowest was in July (58%). During this study, eight aquatic oligochaete taxa were identified, including five tubificines, two rhyacodrilines, and one naidine in the family Naiaidae (Table 3). Species composition, except for June and July 2010, mainly consisted of tubificines and rhyacodrilines, but naidine populations in June and July only accounted for about 5% of the total population.

Limnodrilus hoffmeisteri Claparede, 1862 and B. sowerbyi were observed during all months. In particular, all L. hoffmeisteri had a "plate-topped form" (Brinkhurst and Jemieson, 1971) of the penis sheath.
Seasonal variation in population density

The average annual population density of aquatic oligochaetes was 2,822 m\(^{-2}\) and 24\% (672 m\(^{-2}\)) of the specimens were *B. sowerbyi* (Fig. 3). The highest abundance of 5,836 m\(^{-2}\) occurred in May 2011. Abundance declined continuously from June 2010 to August 2010.

The lowest population density of *B. sowerbyi* was found in June 2010. However, it increased to the average population density of 2010 in July 2010. *B. sowerbyi* remained at the same population density from June 2010 to November 2010 but increased from March 2011 to May 2011. Dominance of *B. sowerbyi* in June 2010 was about 5\%, but increased to approximately 30\% during the summer. In March 2011, *B. sowerbyi* represented more than half of all aquatic oligochaetes.

Seasonal changes in biomass

The average annual biomass of all aquatic oligochaetes was 21.2 g wet wt. m\(^{-2}\) (Fig. 4). Mature *B. sowerbyi* weight was approximately 10 times greater than that of *Limnodrilus* spp. The average biomass of the *B. sowerbyi* population was 66\% (14.2 g wet wt. m\(^{-2}\)) of the total aquatic oligochaete biomass. The *B. sowerbyi* biomass was 91\% of the total aquatic oligochaete biomass in March 2011. Therefore, the total biomass of aquatic oligochaetes reflected changes in *B. sowerbyi*. The largest biomass of all aquatic oligochaetes was 35.0 g wet wt. m\(^{-2}\) in April 2011. However, total biomass in Nov 2010 was high (19.0 g wet wt. m\(^{-2}\)) despite the low biomass of *B. sowerbyi* (6.2 g wet wt. m\(^{-2}\)). This result was reflected in the population density at that time.

Seasonal changes in Branchiura sowerbyi size

The size class distribution of *B. sowerbyi* individuals differed for each month (Fig. 5). The smallest size class (<10 mg) was observed in all months during the study period. This class was most numerous from September 2010 to March 2011 and in May 2011.

The reported minimum breeding size of *B. sowerbyi* is 22 mg (Marchese and Brinkhurst, 1996) or 20–30 mg (Liang, 1984). Ducrot et al. (2007) showed that breeding started at a weight of 84 mg. In contrast, Casellato et al. (1987) showed that the biological cycle was distinguished by seasonal change without relying on individual size by considering the ultrastructural analysis of gonads as well as protogonia and spermatogonia, but this analysis could not be done during this study. Therefore, our study was based on the assumption that *B. sowerbyi* reproduction occurs in worms >20 mg. The >20 mg size class accounted for 35\% of the total annual number of *B. sowerbyi*, and the proportion of this size class decreased consistently from July 2010 to October 2010.
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Fig. 5. Size class distribution of B. sowerbyi at the Kamakura paddy field. Field surveys were conducted once a month between 2010 and 2011, except in the winter (December 2010 to February 2011).

proportion (67%) of the >20 mg class in the population occurred in April 2011. In contrast, the proportion of the smallest size class (<10 mg) increased consistently from July 2010 to October 2010, and again in May 2011.

Discussion

Aquatic oligochaetes are a major component of the invertebrate fauna of paddy field soils (Simpson et al., 1993a, b; Kimura, 2005). The species composition of aquatic oligochaetes in the Kamakura paddy field agreed with the patterns observed in the paddy fields mentioned above (Simpson et al., 1993a, b) and in Lake Kitaura (Ohtaka and Kituchi, 1997). Populations of oligochaetes in paddy soils are often dominated by L. hoffmeisteri and B. sowerbyi (Kikuchi et al., 1975; Simpson et al., 1993a, b). Similar results were found in this study (Table 3). However, our results did not agree with the findings of Yokota and Kaneko (2002), who reported that Dero dorsalis Ferronnière, 1899 were dominant in a paddy in western Japan. Their study showed that a high density of D. dorsalis was recorded in non-ploughed plots with legume mulching. Barton and Farmer (1997) stated that the number of tubificids collected under conventional tillage was increased more than that of collections from conservation tillage. These results suggest that farming method affect the species composition of oligochaetes in paddy fields. In Japanese lakes, Ohtaka et al. (1990) distinguished between two forms of L. hoffmeisteri on the basis of a difference in penis sheaths: the "plate-topped" form is found in eutrophic or organically polluted waters, and the "typical" form is mainly found in oligotrophic, less polluted waters in Japan (Ohtaka et al., 1990). Moreover, B. sowerbyi is very tolerant of low oxygen levels associated with organically polluted waters (Aston, 1984) and Aulodrilus species usually accompany L. hoffmeisteri at a lower density (Ohtaka et al., 1990; Ohtaka and Kituchi, 1997). The dominant aquatic oligochaetes in the Kamakura paddy field were the "plate-topped" form of L. hoffmeisteri and B. sowerbyi, therefore, this study indicates that the Kamakura paddy field is eutrophic. Typically, paddy fields are a type of artificial bog created for agricultural purposes and contain much organic matter with a thick ploughed layer. Therefore, the environmental conditions of paddy fields are similar to those of eutrophic lakes. Moreover, Embolocephalus yamaguchii (Brinkhurst, 1971) was found in the study paddy field. This species was once considered endemic to Lake Biwa (Ohtaka and Nishino, 1995), but has recently been discovered from the intake area and freshwater streams in northern and western Japan (Ohtaka and Martin, 2011) as well as in southern Australia (Pinder and McEvoy, 2002). This is the first time to collect E. yamaguchii from paddy field. The irrigation water of the Kamakura paddy field originates from reservoirs and is supported by spring water from the surrounding hills. Therefore, E. yamaguchii seems to be supplied from stream water.

Population density in Kamakura was lower (in the middle
of July and at the end of the crop season population density of aquatic oligochaetes was 1,592 m\(^{-2}\) and 4,633 m\(^{-2}\), respectively) than in the paddy fields mentioned above and a lake with a similar species composition was reported. The population density estimated in northern Japan (Kikuchi et al., 1975) was 50,000 m\(^{-2}\) in the middle of July where combined densities of L. hoffmeisteri and B. sowerbyi, and the Philippines amount of oligochaete population densities (Simpson et al., 1993b) was 8,200 m\(^{-2}\) (ranged from 0 to 35,000 m\(^{-2}\)) at the end of the crop season. In Lake Kitaura, a eutrophic lake in central Japan, during the period from 1981 to 1984, the annual density of the most dominant oligochaete, L. hoffmeisteri ranged from 27,600 m\(^{-2}\) to 7,850 m\(^{-2}\) without any seasonal trend. Among other oligochaetes (in 9 species including B. sowerbyi), only three species exceeded 600 m\(^{-2}\) in some sampling occasions (Ohtaka and Kikuchi, 1997). On the other hand, similar values of 2,500 m\(^{-2}\) have been reported in western Japan, where a dissimilar species composition dominated by D. dorsalis was observed at the end of the crop season (Yokota and Kaneko, 2002).

B. sowerbyi showed strong seasonal changes in its life cycle. In our case, this pattern is related to seasonal changes in temperature (Fig. 5) and nutrients (Table 2). Previous studies have shown that the life history of B. sowerbyi is normally limited by temperature and the quantity and quality of food (Casellato, 1984; Clark et al., 1989; Bonacina et al., 1994; Raburu et al., 2002; Nascimento and Alves, 2008). Aston et al., (1982) reported that higher growth rates occurred at 21–29 °C the highest growth rates occurred at 25°C, the highest mortalities occurred at temperatures >29°C, and low growth rates occurred at >33°C and <17°C. Hatching from cocoons starts at 15°C, and reaches its maximum at 25°C. Hatching decreases when temperatures are lower or higher (Aston et al., 1982; Bonacina et al., 1994). No cocoons were laid at temperatures <17°C (Aston et al., 1982). At the study site, the monthly average soil temperature was approximately 25 °C from June to September, 18°C in October, and 12°C in November. The temperature gradually increased until May 2011 after a decline to near 0°C in January 2011 (Fig. 2). In this study, growth and reproduction of B. sowerbyi were optimal from June to September, and population changes were assumed to be small from November 2010 to March 2011, because of the stable continuous water supply (winter flooding) and low temperature. Moreover, growth and reproduction occurred in each size cohort starting in April 2011. Because the worms started laying eggs in May, the <10 mg cohort of individuals in June were newborn. Growth and reproduction continued until September and declined in October; thus, B. sowerbyi observed in April had probably overwintered. Temperature increased from April through May, when reproduction starts, and the population of <10 mg-sized individuals increased in May. In addition, seasonal changes in the life cycle might be influenced by the addition of plant residues. The nitrogen and carbon concentrations were high after harvest (Table 2). In Lake Naivasha, aquatic oligochaete biomass peaks were synchronized with increases in food resource (Raburu et al., 2002). It might be possible that individual worms were able to grow in autumn, because the nitrogen and carbon concentrations increased after harvest when plant residues were left in the paddy.

The population dynamics of aquatic oligochaetes may be affected by human disturbance (Table 1). Floodwater management is the most important determinant of aquatic oligochaete population dynamics (Simpson et al., 1993b). The loss of water imposes a potential catastrophe on aquatic fauna, because many species cannot adapt to or escape from the dry condition (Wissinger, 1999; Tarr et al., 2005). The absence of water in paddy fields during tillage suppresses the occurrence and density of some aquatic macroinvertebrate taxa with a low tolerance to drought (Stenert et al., 2009). Winter flooding in paddy fields sustains high levels of biological diversity, which is important from a conservation standpoint. Aquatic oligochaetes can survive during winter due to winter flooding; therefore, our results suggest that aquatic oligochaetes were able to maintain their numbers until spring. Tillage practises are considered to be important for soil management but they physically disturb paddy soils. Soil puddling was performed in June at the study field using a rotary machine which disturbed the upper 20 cm of soil. Our results suggest that tillage had a minor effect on the total population of aquatic oligochaetes in June. The population density of B. sowerbyi decreased temporarily (Fig. 3), perhaps because the soil puddling reduced their numbers through mechanical cutting and because of low levels of dissolved oxygen resulting from consumption by microorganisms. Soil analysis showed that soil carbon content was not significantly different from June to November after soil puddling, suggesting that tillage itself affected B. sowerbyi. Yokota and Kaneko (2002) indicated that aquatic oligochaetes take 69 days to recover under non-tillage conditions and 90 days to recover from ploughing treatment under tillage management after irrigation. In this study, B. sowerbyi recovered after 30 days, and large individuals were
collected in July. At the time of soil palling, *B. sowerbyi* may have escaped to a deeper layer and started reproducing. These results suggest that decreases in population density caused by soil palling can be reversed.

No stress, such as pesticides or drought, was present in the Kamakura paddy field. Therefore, population dynamics of aquatic oligochaetes were related to changes in environmental and management conditions of the paddy field. In a previous study, the quality and quantity of food appeared to be the major limiting factors in the population dynamics of benthic macro-invertebrates in Lake Naivasha (Raburu et al., 2002). However, our results suggest that in addition to food quantity, seasonal changes in temperature have an important effect on the populations of aquatic oligochaetes in the shallow artificial wetland environment of an organic farming paddy field.

Data on seasonal changes in species composition, population density, and biomass of aquatic oligochaetes are necessary to assess ecosystem function. Further research is necessary to clarify the ecosystem function of aquatic oligochaetes in organic agriculture. Therefore, further research needs to study the effects of differences in species composition, population density, and biomass of aquatic oligochaetes on function in the sediment. These studies will allow the assessment of ecological risks, for example, when non-target aquatic oligochaetes are lost as a result of the use of agricultural chemicals.

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**References**


Community structure of aquatic oligochaetes in an organic paddy


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