A comparison of predacious aquatic insect fauna and density in ground pools and concrete pools created during the Great East Japan Earthquake in 2011

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Abstract: The tsunami caused by the Great East Japan Earthquake on March 11, 2011 hit the Pacific coast and caused heavy destruction of natural and man-made environments in north-eastern Japan. This study focuses on mosquito larvae and their potential aquatic insect predators associated with ground pools and pools that appeared in the concrete foundations of destroyed houses (concrete pool) in inundated areas in Miyagi Prefecture, Japan. Field samplings were conducted on late July 2013. Culex inatomii, Cx. pipiens group, Cx. tritaeniorhynchus, and Cx. orientalis were collected from ground pools and concrete pools. The abundance of Cx. inatomii and Cx. pipiens groups in concrete pools was significantly greater than that in the ground pools. A large number of Hydroglyphus japonicus were collected as potential mosquito predators, followed by Micronecta spp., Enochrus japonicus, Rhantus sturalis, Aquarius paludm paludum and Hydrochora affinis, categorized as “flight dispersers,” which might immigrate rapidly from the non-inundated rice fields or wetlands. Stepwise generalized linear models suggested that larval abundance of Cx. inatomii in the pools studied was affected by the vegetation cover and habitat type (ground pool or concrete pool), but not by water depth, salinity, presence of predators, and bottom type (sand or concrete) of aquatic bodies. Concrete pools and covered with dense vegetation provide breeding habitat for Cx. inatomii along with their potential predators.

Key words: aquatic insects, biocommunity, brackish water, dipper, dipnet, disaster, tsunami

INTRODUCTION

On March 11, 2011 a tsunami caused by the Great East Japan Earthquake hit the Pacific coast of north-eastern Japan. The tsunami completely destroyed urban and rural environments, including buildings, roads, bridges, agricultural fields, and natural vegetation. Abandoned paddy fields, destroyed houses, and ground pools were found containing brackish water carried by the tsunami (Kobayashi et al., 2012; Tsuda et al., 2012; Watanabe et al., 2012). These brackish water pools provide larval habitats of vector mosquitoes and need to be monitored to evaluate risk for outbreak of mosquito-borne disease. These brackish water pools were utilized by potential West Nile vector mosquito, Culex inatomii (Kamimura and Wada), Culex pipiens group, and Culex tritaeniorhynchus Giles, as larval habitats, suggesting the expansion of the distribution ranges of these mosquito species in 2011 (Tsuda et al., 2012; Watanabe et al., 2012; Tsuda and Kim, 2013). The adult mosquito density in 2012 was lower than that in 2011; however, Cx. pipiens group and Cx. inatomii reached a high density in the summer of 2012, indicating that the outbreaks of these mosquitoes occurred again in tsunami areas in 2012 (Tsuda et al., 2013).

The impact of natural predators on the survival of mosquito larvae has been evaluated in several ecological studies in wetlands such as rice agroecosystems (Service, 1977; Mogi and Miyagi, 1990; Mogi, 1993; Takagi et al., 1996; Shaalan and Canyon, 2009; Ohba et al., 2013). Because tsunamis are destructive and rare natural events, our knowledge of their ecological impacts on inland water ecosystems is limited (Sites and Vitheepradit, 2010). According to Sites and Vitheepradit (2010), the ponds inundated by the tsunami caused by the Sumatra–Andaman earthquake in 2004 were rapidly colonized by taxa with high salt tolerance. Mukai et al. (2014) studied the ecological impacts of the tsunami caused by the Great East Japan Earthquake on aquatic animals in rice paddies in the Tohoku area in 2012. The total number of walk dispersers (frogs) and obligatory aquatic animals without resting stages (Mollusca and fish) were significantly smaller in inundated than in non-inundated rice paddies, but no notable difference was detected between the rice paddies in taxonomic richness and the total number of observations of flight dispersers (insects) and obligatory aquatic animals with resting stages (crustaceans) (Mukai et al., 2014). However, little research has been devoted to aquatic animals such as mosquito predators in areas destroyed...
This study focuses on mosquitoes and their potential aquatic insect predators associated with ground pools and pools in the concrete foundations of destroyed houses (concrete pools) in inundated areas in Miyagi Prefecture. Because information about the colonization by aquatic insect predators in the concrete pools is limited, this study will provide ecological notes on these aquatic insects.

**Materials and Methods**

**Study area**

Field collections to examine aquatic animals in eastern Miyagi Prefecture, Tohoku, Japan (38°09′ N, 140°56′ E; Fig. 1) were conducted between July 29 and 31, 2013 (Fig. 1). This period was within the rainy season and mosquito abundance was high in this area in 2011 and 2012 (Tsuda et al., 2012; Tsuda et al., 2013). A tsunami, caused by the earthquake on March 11, 2011, reached inland 5–6 km from the seashore and destroyed these study areas, including buildings, dwellings, roads, as well as agricultural fields and natural vegetation. There were many ground pools of various sizes containing brackish water. Twenty-three ground pools (>1.0 m² in water area) in an area east of Sendai Airport (Natori City), and 13 pools (>0.8 m² in water area) in the concrete foundations of houses destroyed (concrete pools) by the tsunami in Wakabayashi-Ku, Sendai City were selected as study plots. These areas are within 0.5 km from the seashore (Fig. 1).

Salinity, conductivity, TDS, and pH in each pool were measured using a digital salinity meter (SK-5SII; Sato Keiryoki MFG. Co., Ltd.) and digital testers (Dist5; Hannna Instruments Co., Ltd.). The vegetation covering pools was roughly estimated using photos taken by a digital camera (FinePix XP200, Fujifilm Corporation, Tokyo) (score 1=0–20%, 2=21–40%, 3=41–60%, 4=61–80%, 5=81–100% cover). The bottom type of pools (sand/mud or concrete) and the presence of floating leaves along with emergent and submerged plants (positive or negative) in each pool was also confirmed by a visual inspection.

**Sampling methods**

We monitored the abundance of mosquito larvae and other insects using a dipping method as described in Ohba et al. (2013). The dipper used for collection was 12 cm in diameter and 5 cm deep. We collected one sample from each study pool; each sample consisted of 30 dips made at 30 different sites with >1 m separation between them. When it was impossible to perform 30 dips due to the small size of an aquatic habitat, we made 10 or 20 dips and extrapolated the number to 30 dips. In addition to the dipping method, a sweeping census using a 1-mm-mesh D-frame dipnet (0.28 m wide) was performed for 5–10 minutes in order to investigate aquatic insects that were difficult to catch using the dipping method. Collected aquatic insects were preserved in 80% ethanol for later identification. Insects, excluding mosquito larvae, were identified to the order, family, genus, or species level using a binocular microscope and reference (Mori and Kitayama, 2002; Kawai and Tanida, 2005). On the basis of the literature (Mogi, 2007; Alahmed et al., 2009; Shaalan and Canyon, 2009), Coleoptera, Heteroptera and Odonata were regarded as potential insect predators of mosquito. All mosquito larvae, excluding damaged and/or first- to third-instar larvae, were identified using taxonomic keys (Tanaka et al., 1979). Pupae were reared until emergence for the purpose of species identification. Larvae smaller than the fourth instar were identified to the genus level and counted.
Data analysis

One-way ANOVA was used to compare aquatic animal abundance in ground pools and concrete pools. Log_{10} (n + 0.5) transformation of exact values of aquatic animal abundance was performed to standardize variances and improve normality, in order to satisfy the assumptions of the ANOVA model (Yamamura, 1999). The proportion of floating leaves and emergent and submerged plants, and vegetation cover rate were compared using the chi-square test and Mann–Whitney U test, respectively.

* Culex inatomii were the most abundant mosquito species in the study site (see Results). Although the immature development in this species was affected by the concentration of salinity water, ecological factors such as predator and vegetation which determine the larval abundance of *Cx. inatomii* have never been evaluated in the field (Katano et al., 2010). We used a general linear model (GLM) with negative binomial (“glm.nb” in MASS package) in R version 3.1.2 (R Core Team, 2014) to determine *Cx. inatomii* abundance. Habitat (ground pool or concrete pool), presence of predators (presence or absence; evaluated by both dipper and dip net), water depth, salinity, vegetation cover rate, and bottom type (sand or concrete) were the factors used in the analysis. We did not use “ predator abundance” as explanatory variables in GLM because some potential insect predators of mosquito were not captured and not evaluated its exact abundance by the dipper method (see Table 2). In the preliminary analysis, second order interactions concerning presence of predators, habitat, and bottom type were not significant. Therefore, we did not include any interaction terms in the GLM. The backward stepwise method using the stepAIC package was used for model selection.

**Results and Discussion**

Generally, as time progressed, the salinity attenuated due to rain water, leading to dense growth of aquatic plants. Mean salinity in both the ground pools and concrete pools was 0.02% (ranging from 0.00% to 0.06% in both pool types) (Table 1). Water depth and pH in the concrete pools were significantly larger than those in the ground pools (Table 1). The percentages of emergent and submerged plants and vegetation cover in ground pools were statistically greater than those in the concrete pools (Table 1).

Four species of *Culex* were collected in this study from both aquatic habitats: 221 *Cx. inatomii*, 18 *Cx. pipiens* respectively.

| Species          | Ground pool (23) | Concrete pool (13) | P   |
|------------------|------------------|--------------------|-----|
| *Cx. inatomii*   | 1.65±2.74        | 14.08±21.43        | 0.029 |
| *Cx. pipiens gr.*| 0.00±0.00        | 1.38±2.84          | <0.001 |
| *Cx. tritaeniorhynchus* | 0.17±0.65   | 0.00±0.00          | 0.307 |
| *Cx. orientalis* | 0.78±1.81        | 0.00±0.00          | 0.119 |

*ANOVA was performed after log (n + 0.5) transformation.*
group, 4 Cx. tritaeniorhynchus, and 18 Cx. orientalis Edwards. The larval densities of Cx. inatomii and Cx. pipiens group in concrete pools were significantly greater than those in the ground pools (Table 2). No larvae of Cx. tritaeniorhynchus or Cx. orientalis were found in concrete pools, and no larvae of Cx. pipiens group were found in the ground pools. Culex inatomii larvae were found in 39.1% of ground pools and 46.2% of concrete pools. Culex pipiens group larvae were found in 38.4% of concrete pools.

In terms of potential insect predators of mosquito, 281 individuals, including 5 hemipterans and 10 coleopterans, were collected from both aquatic habitats. A large number of Hydroglyphus japonicus (Sharp) were collected, followed by Micronecta spp., Enochrus japonicus (Sharp), Rhantus suturalis (Macleay), Aquarius paludim paludum (Fabricius), and Hydrochara affinis (Sharp) (Table 3). These species are categorized as “flight dispersers” as they immigrate rapidly from the non-inundated rice fields or wetlands (Mukai et al., 2014). The numbers of H. japonicus and A. paludim paludum in concrete pools were significantly higher than in the ground pools (Fig. 2). Although the reason for this difference is unclear at present, these species have high adaptive ability to various aquatic environments, such as pools surrounded by hard concrete (Mori and Kitayama, 2002), and prefer open water (Negoro, 1988).

In concrete pools, larvae of E. japonicus, H. japonicus, R. suturalis and nymphs of A. paludim paludum were found, indicating they have breeding populations in these pools. Larvae (prepupa) and newly emerged adults of R. suturalis were found under the soils (Fig. 3). Because R. suturalis is expected to be an efficient predator of mosquito larva (Ohba and Takagi, 2010), this “aquatic-terrestrial ecotone” may provide good breeding sites for the species. In addition, Allodessus megacephalus (Gschwendtner) was found in pools in the concrete pools. This habitat may be similar to the primary habitat of this species (Mori and Kitayama, 2002); L. megacephalus inhabits brackish water such as tidal pools near the coastline. In the preliminary survey in June 2013 (data not shown), we confirmed one Hygrotus chinensis (Sharp) in ground pools in this study site. This species is listed as “Vulnerable” in the Red List.
of Miyagi Prefecture (Association of Wildlife Research & EnVision, 2013) and ecological notes on this species in Japan are limited.

Stepwise GLMs (with negative binomial) revealed that Cx. inatomii larval abundance in the pools studied was affected by the vegetation cover rate and habitat (ground pools or concrete pools), but not by water depth (Table 4). Salinity, presence of predators (presence or absence), and bottom type (sand or concrete) were excluded from the first model in the stepwise process. An increase in vegetation cover rate would provide a larval habitat for Cx. inatomii. The relationship between larval density of Cx. inatomii and vegetation density or cover rate was hardly revealed. According to Tsuda (2013), his observation suggested that the larvae were found in sunny pools of sparsely reed bed. For the ground pools, the vegetation cover rate was decreased by periodical weeding and the suitable mosquito habitats were gradually decreased: 18.5% (60/324 sites) in 2012 and 8.9% (15/168 sites) in 2013 (Y. Tsuda unpubl. data). Hence, concrete pools in this study site will become larval habitats for Cx. inatomii.

Predators (Coleoptera and Hemiptera) did not affect the density of Cx. inatomii larvae (Table 4). Although we did not confirm if these predators feed on mosquitoes and no significant relationship was found between their presence and mosquito abundance in this study, it is likely that they consume mosquito larvae (Mogi, 2007; Shaalan and Canyon, 2009). In previous studies, one-time samplings from a variety of aquatic habitats did not detect the effect of predators on mosquito abundance (Ohba et al., 2011; Ohba et al., 2015), but repeated samplings from certain aquatic habitats during a long period (5 months) could detect the negative predator effect on mosquito abundance (Ohba et al., 2013). Therefore, repeated sampling is necessary to reveal predator–mosquito relationships. In addition, Culex tritaeniorynchus (Ohba et al., 2012) and Cx. pipiens pipiens (S. Ohba unpubl. data) larvae display motionless behavior at the water surface when they detect predator cue, being a form of predator avoidance. Assuming that Cx. inatomii have this predator avoidance ability, presence of predator might not affect the density or presence of Cx. inatomii. Further studies should evaluate the behavioral change of Cx. inatomii when they are exposed to predator cue.

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| Source | Parameter estimate | S.E. | Z | P  |
|--------|--------------------|------|---|----|
| Intercept | 2.381 | 0.819 | 2.908 | 0.0036** |
| Water depth | −0.061 | 0.033 | −1.835 | 0.0666 |
| Vegetation density | 0.990 | 0.365 | 2.710 | 0.0067** |
| Habitat† | −3.732 | 0.856 | −4.359 | <0.0001*** |

†The coefficient indicates the relative effect of ground pool compared with pool in house concrete foundations.
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