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After-effects of Tsunami on distribution and abundance of mosquitoes in rice-field areas in Miyagi Prefecture, Japan in 2011

Yoshio Tsuda*[, 1)], Michio Haseyama[2)], Keiichi Ishida[3)], Jun Niizuma[4)], Kyeong Soon Kim[1)], Daiki Yanagi[2)], Nozomi Watanabe[2)] and Mutsuo Kobayashi[1)]

* *Corresponding author: Department of Medical Entomology, National Institute of Infectious Diseases, Toyama 1–23–1, Shinjuku-ku, Tokyo, 162–8640 Japan*

[1)] *Department of Medical Entomology, National Institute of Infectious Diseases, Toyama 1–23–1, Shinjuku-ku, Tokyo, 162–8640 Japan*

[2)] *Narita Airport Quarantine Station, Aza-Furugome 1–1, Furugome, Narita, Chiba, 282–0004 Japan*

[3)] *Sendai Quarantine Station, Teizandori 3–4–1, Shiogama, Miyagi, 985–0011 Japan*

[4)] *Tokyo Quarantine Station, Aomi 2–7–11, Koto-ku, Tokyo, 135–0064 Japan*

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Abstract: The Tsunami caused by the Great East Japan Earthquake on 11 March 2011 in north-eastern Japan destroyed urban and rural environments completely, including buildings, dwellings, roads, bridges, agricultural fields and natural vegetation. The after-effects of the Tsunami on the distribution and abundance of mosquitoes were studied in rice field areas in Miyagi Prefecture, Japan during June to August 2011. Adult mosquitoes were collected by traps enhanced with 1 kg dry ice and the density was compared between the "Tsunani" area and "No Tsunami" area. A total of 6,542 adults of 9 species in 4 genera were collected and the following 4 species were dominant: *Culex pipiens pallens, Cx. inatomii, Cx. tritaeniorhynchus* and *Aedes albopictus*. The high density and expanded distribution of *Cx. inatomii* was marked. The adult density of the former 3 dominant species was significantly higher in the Tsunami area than No Tsunami area, while *Ae. albopictus* was significantly abundant in the No Tsunami area. Ground pools in disaster areas contained brackish water with 0.47 and 0.21% average salinity in June and August 2011, respectively. The percentage of water samples with mosquito larvae increased from 2.7% in June to 79.5% in August, suggesting the expansion of the larval distribution of *Cx. inatomii* and *Cx. tritaeniorhynchus* as well as *Anopheles sinensis*-group.

Key words: Tsunami, earthquake, north eastern Japan, mosquitoes, *Culex inatomii*

Introduction

The disaster caused by the Great East Japan Earthquake and Tsunami on 11 March 2011 in north-eastern Japan was serious, and human dwellings, buildings, roads, bridges and agricultural fields as well as natural vegetation were destroyed completely both in urban and rural areas. Because of the shortage of energy and manpower, most activities to recover from the damage have been concentrated in urban areas. The recovery of destroyed agricultural fields has progressed slowly and most of the destroyed fields in rural areas have been remained as they are.

The main disaster area is roughly divided into two geographically different areas: hilly areas with a deeply indented coastline that are located mainly in Iwate Prefecture and northern Miyagi Prefecture; flat areas with a smooth coastline that are located from southern Miyagi Prefecture to Fukushima Prefecture. The flat areas are used for rice plantation and rice paddies are distributed widely. Rice fields are the major larval habitats of mosquitoes of medical importance in Japan, such as *Culex tritaeniorhynchus* Giles, *Anopheles sinensis* Wiedemann and *Aedes vexans nipponii*

(Theobald). Kamimura (1968) collected 17, 24, and 25 mosquito species in Iwate, Miyagi and Fukushima Prefecture, respectively, in the 1960s. Regarding rice field mosquitoes in Miyagi Prefecture, Kato et al. (1967) conducted detailed studies on the seasonal abundance of mosquitoes in cow sheds located in 14 study areas and the following 5 mosquito species were dominant in 1965, *An. sinensis*, *Cx. tritaeniorhynchus, Ae. vexans nipponii*, *Armigeres subalbatus* (Coquillett), *Cx. pipiens pallens* Coquillett. No data are available regarding the recent mosquito situation in disaster areas.

The destruction of the environment by the earthquake and Tsunami is so complete and the disaster area is so wide that the recovery of populations of wild animals and plants will progress gradually each year. This is an important opportunity not only scientifically but also practically to apply ecological theories and predict the recovery process of wild populations. We selected rice field areas in Miyagi Prefecture for mosquito surveys in this study to clarify the dominant species and elucidate the after-effects of a Tsunami on the distribution and abundance of mosquitoes in disaster areas.

Materials and Methods

Study area: The disaster area is roughly divided into two areas based on the geographical features; hilly areas with a deeply indented coastline and flat areas with a smooth coastline. We examined the flat areas where rice plants were cultivated widely located in the southern part of Miyagi Prefecture, Japan (Fig. 1). A huge Tsunami, caused by the earthquake on 11 March 2011, reached regions 5 to 6 km from the seashore. Almost all human dwellings and buildings along the seashore were completely destroyed and

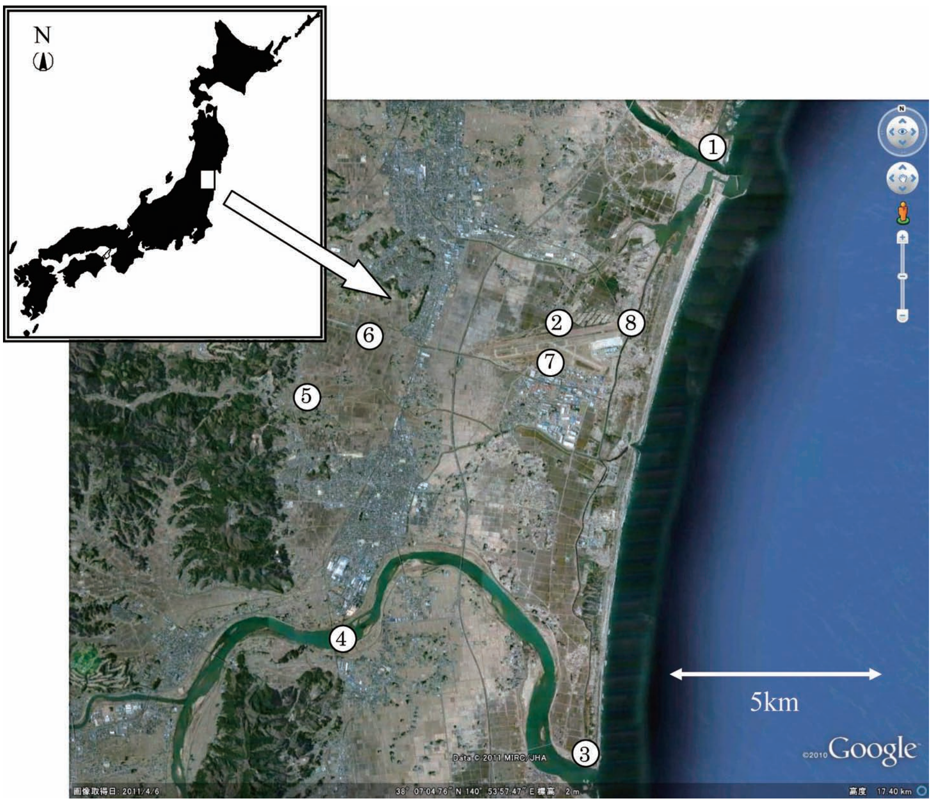

Fig. 1. Locations of 8 sites for adult mosquito collections in rice-field areas in Miyagi Prefecture, Japan. Two CO_2-baited traps were placed at approximately 10 m distance at each site. See Table 1 for details.

washed away. In rice field areas, agricultural fields were flooded with seawater and damaged heavily by debris carried by the Tsunami. There were many ground pools of various sizes containing brackish water (Fig. 2).

Within the study area we could easily distinguish two areas, either damaged or undamaged by the Tsunami, and the boundary was about 5 to 6 km from the seashore. Mosquito surveys were conducted in both the damaged "Tsunami" area, and undamaged "No Tsunami" area to compare the distribution and abundance of mosquitoes.

Mosquito survey: Mosquito surveys were carried out monthly from June to August 2011. Both adult and larval collections were conducted. For adult collections, 10 CDC-like traps without a bulb baited with 1 kg dry ice were used and operated continuously for 3 days. Mosquitoes in the traps were collected every morning and taken to Sendai Quarantine Station for species identification. Mosquito species was identified following the morphological keys by Tanaka et al. (1979). For the identification of *Cx. pipiens* groups, the PCR-based method developed by Kasai et al. (2008) was used, and 28 adults collected in June, July and August 2011 were identified. Since all the specimens were identified as *Cx. pipiens pallens*, the remaining samples were treated as *Cx. pipiens pallens* in this paper. Five trap sites (1 to 5 in Fig. 1) were selected, and 2 traps each were placed and operated for 3, 2 and 1 day(s) in June, July and August 2011, respectively. In July and August, the trap sites were selected along a transect from the seashore to inland (2, 5, 6, 7 and 8 in Fig. 1) and adult collections were conducted by using 2 traps each for 1 and 2 days, respec-

Fig. 2. (a) A ground pool in a depression along the roadside on 21 July 2011, (b) a ground pool in a destroyed green house, on 21 July 2011, (c) rice fields with brackish water on 14 June 2011, (d) grasses grew in rice fields on 30 August 2011 and ground pools became suitable larval habitats for *Cx. inatomii* and *Cx. tritaeniorhynchus*.

Table 1. Location and environmental conditions of mosquito-trap sites in rice-field areas of Miyagi Prefecture, Japan.

	Location	Distance from seashore, km	Environmental conditions
1	N38.10.56, E140.57.23	0.5	Tsunami area, river mouth of Natori river
2	N38.8.31.896, E140.54.53.886	2.6	Tsunami area, north of Sendai airport
3	N38.3.8.115, E140.55.7.153	0.25	Tsunami area, river mouth of Abukuma river
4	N38.5.33, E140.52.15	5.0	No Tsunami area, riverside bush along Abukuma river
5	N38.8.7.624, E140.50.45.284	8.3	No Tsunami area, beside a rural house
6	N38.8.36.73, E140.51.54.91	6.2	No Tsunami area, center of a rice-field area
7	N38.8.31.267 E140.54.51.279	2.5	Tsunami area, south of Sendai airport
8	N38.9.5 E140.56.29	0.9	Tsunami area, east of Sendai airport

tively, to show the different spatial distribution of host-seeking females among mosquito species. The location and environmental conditions of the trap sites are listed in Table 1.

Collection sites of larvae were selected from both the Tsunami area and No Tsunami area. Rice paddies with growing rice plants in the No Tsunami area were examined by a dipper (10 cm diameter and 350 m*l*) and 38, 12 and 9 samples consisting of 5 to 10 dips of water were collected in June, July and August 2011. A total of 37, 26 and 35 larval samples were collected from ground pools found in the Tsunami area in June, July and August 2011, respectively. Salinity of the collected water was measured by a digital salinity meter (SS-31A; Sekisui) and recorded in the fields. Larvae were killed and kept in alcohol for later identification. Larvae of *Anopheles sinensis* Wiedemann, *An. sineroides* Yamada, and *An. lesteri* Baisas and Hu are almost indistinguishable (Tanaka et al., 1979) and most of our larval samples included young instars, therefore these *Anopheles* larvae were treated as *Anopheles sinensis*-group (Tanaka, 2005).

Blood-meal identification: Seven and one blood fed-females of *Cx. pipiens* group and *Cx. inatomii* Kamimura and Wada, respectively, were collected by dry-ice traps and blood-meal identification was carried out following Kim et al. (2009a).

Statistical analysis: Average number of females/trap/day was calculated for dominant mosquito species and compared between Tsunami and No Tsunami areas. The significance of the observed difference in mosquito density was tested by t-test by applying log $(1+n)$ transformation. To describe the spatial distribution of host-seeking females along a transect from the seashore to inland, the relationships between the distance from seashore and average numbers of females/trap/day were analyzed by exponential curve fitting.

Results

Total of 6,542 adult mosquitoes of 9 species in 4 genera were collected during the study period (Table 2). A large number of *Culex inatomii* were collected as well as *Cx. pipiens pallens*, *Cx. tritaeniorhynchus* and *Aedes albopictus* (Skuse). The remaining 6 species, *Armigeres subalbatus*, *Cx. orientalis* Edwards, *An. sinensis*, *Cx. bitaeniorhynchus* Giles and *Ae. vexans nipponii,* were collected in small numbers, less than 10 in total.

Statistically significant differences in the average number of females were found between Tsunami and No Tsunami areas for 4 dominant species in July and August 2011 (Table 3). Density of *Cx. pipiens pallens*, *Cx. inatomii* and *Cx. tritaeniorhynchus* in the Tsunami area was significantly higher than in the No Tsunami area, while the opposite result was found for *Ae. albopictus*, which showed higher density in the No Tsunami area than Tsunami area.

The spatial distributions of dominant species are shown in Fig. 3. The exponential curve was fitted well to the relationship be-

Table 2. Results of dry-ice trap collections conducted in Tsunami and No Tsunami areas in rice-field areas in Miyagi Prefecture, Japan during June to August 2011.

Species	No Tsunami area				Tsunami area			
	June (8)	July (12)	August (12)	Total (32)	June (12)	July (18)	August (18)	Total (48)
Cx. pipiens pallens	1	194	133	328	10	2,565	1,415	3,990
Cx. inatomii	0	1	1	2	7	248	1,230	1,485
Cx. tritaeniorhynchus	0	11	86	97	0	86	961	1,047
Ae. albopictus	2	35	249	286	0	1	14	15
Ar. subalbatus	0	1	4	5	0	0	1	1
Cx. orientalis	0	0	2	2	2	0	0	2
An. sinensis	0	1	0	1	0	2	0	2
Cx. bitaeniorhynchus	0	0	1	1	0	0	0	0
Ae. vexans nipponii	1	0	0	1	0	0	0	0
Total	4	243	476	723	19	2,902	3,621	6,542

Number in parentheses shows the total number of trap nights.

Table 3. Comparisons of adult densities (females/trap/day) of four dominant mosquitoes between Tsunami and No Tsunami areas in Miyagi Prefecture, Japan in July and August 2011.

	July			August		
	No Tsunami	Tsunami	p*	No Tsunami	Tsunami	p
Cx. pipiens pallens	22.8 ± 6.9	210.9 ± 69.1	0.005	11.3 ± 13.5	78.5 ± 65.6	0.012
Cx. inatomii	0.1 ± 0.4	20.5 ± 15.4	< 0.001	0.1 ± 0.3	68.3 ± 60.6	< 0.001
Cx. tritaeniorhynchus	1.3 ± 1.0	7.2 ± 5.2	< 0.001	7.2 ± 10.5	53.4 ± 97.2	< 0.001
Ae. albopictus	4.3 ± 8.3	0.1 ± 0.3	0.029	21.3 ± 42.6	0.2 ± 0.5	0.010

*t-test was performed after log $(n+1)$ transformation.

tween the distance from the seashore and the density of females of *Cx. pipiens pallens*, *Cx. inatomii* and *Ae. albopictus*, with coefficients of determination of 0.836, 0.847 and 0.789, respectively. Based on the exponential curves, the density of host-seeking mosquitoes at the boundary between Tsunami and No Tsunami areas was estimated as 26, 1.7 and 1.5 for *Cx. pipiens pallens*, *Cx. inatomii* and *Ae. albopictus*, respectively. The spatial distribution of *Cx. tritaeniorhynchus* showed no clear tendency along the transect from the seashore to inland.

Seven blood-fed *Cx. pipiens pallens*, 5 in July and 2 in August, and 1 blood-fed *Cx. inatomii* in August were collected by dry-ice traps during the study period. The blood source animals were identified as *Parus major* for 5 blood-fed *Cx. pipiens pallens* and *Rattus norvegicus* for *Cx. inatomii*. The remaining 2 samples showed no amplification.

The results of larval surveys are summarized in Table 4. The percentage of water samples containing mosquito larvae was low (2.7%) in June, increased to 31.6% in July and reached 79.5% in August 2011. The results suggested the expansion of the larval distribution of *Cx. inatomii*, *Cx. tritaeniorhynchus* and *An. sinensis*-group during the study period. Although a high adult density of *Cx. pipiens pallens* was observed, their larvae were collected in only 3 samples in August 2011. The density of larvae (number of larvae/dip) observed in August varied from 0.01 to 1.5 depending on species; the average larval density of *Cx. inatomii*, *Cx. tritaeniorhynchus*, *Cx. pipiens pallens*, *Cx. orientalis*, *An. sinensis*-group was 1.5, 0.9, 0.5, 0.02, and 0.01, respectively.

The salinity of collected water ranged be-

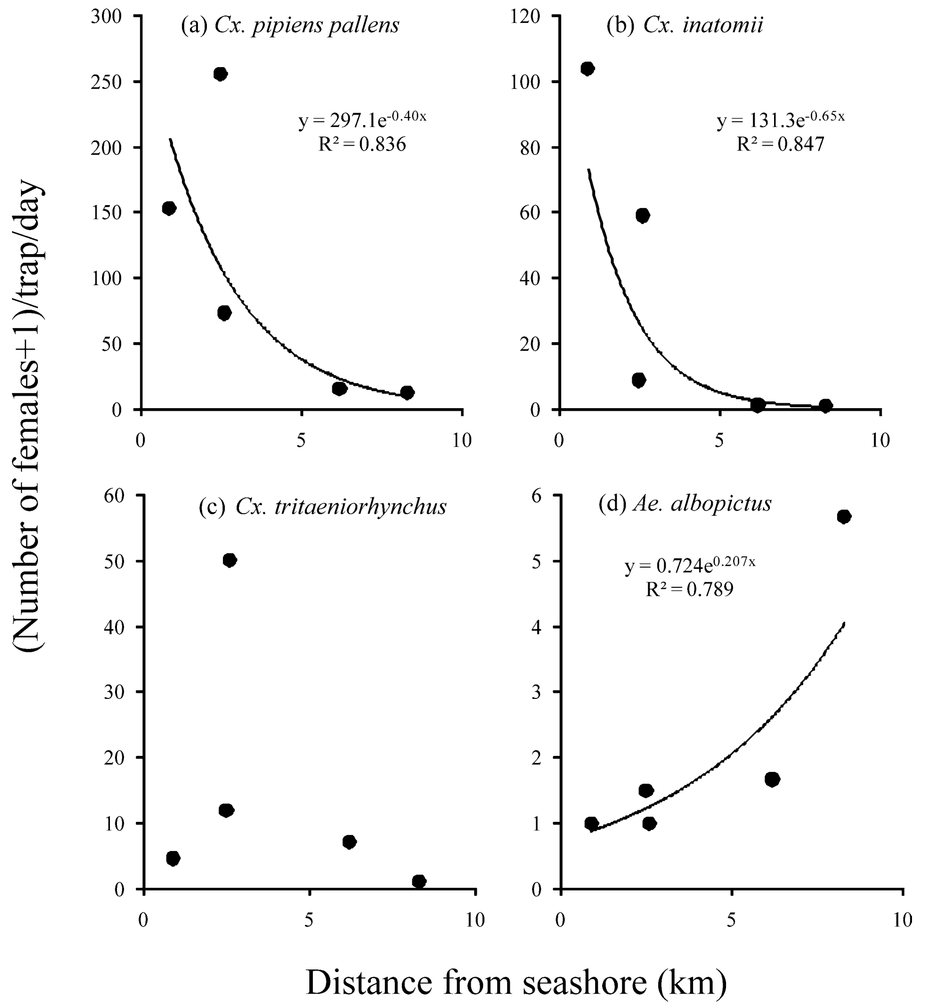

Fig. 3. Relationship between the distance from the seashore and the density of females (1 + *n*/trap/day) for four dominant species observed in Miyagi Prefecture, Japan in 2011.Exponential curve fitting was performed.

tween 0 to 0.9%. The average salinity of sampled water was calculated as 0.47 ± 0.21%, 0.27 ± 0.15% and 0.21 ± 0.19% in June, July and August 2011, respectively. No mosquito larvae were collected from ground pools of 0.8 and 0.9% salinity. Average salinity of water where mosquito larvae were collected was calculated for each mosquito species in Table 5. The average salinity ranged between 0 and 0.33, and differences in average salinity among mosquito species were not statistically significant ($F = 0.289$, $p = 0.755$ in July and $F = 2.024$, $p = 0.111$ in August 2011, ANOVA).

Discussion

The present study clearly showed the aftereffects of the Tsunami on mosquito distribution and abundance in the disaster areas. The high density and expanding distribution of *Cx. inatomii* was marked. *Culex inatomii* had been collected from a few locations in Japan before 2000, such as Okayama and Osaka (Kamimura and Wada, 1974; Mizuta et al., 1999); however, a recent entomological study found this species inhabiting Tokyo bay, Sakata wetlands in Niigata, and Kushiro (Tsuda et al., 2009; Kim et al., 2009b; Katano et al., 2010; Ejiri et al., 2011). A wide geo-

Table 4. Mosquito species found in the larval surveys and the number of samples with larvae collected from rice paddies and ground pools in Miyagi Prefecture, Japan from June to August 2011.

Species	June	July	August
Cx. inatomii	0	2	21
Cx. tritaeniorhynchus	0	7	10
An. sinensis-group	1	4	5
Cx. orientalis	1	0	4
Cx. pseudovishnui	1	0	0
Cx. pipiens pallens	0	0	3
Number of samples with mosquito larvae (%)	2 (2.7)	12 (31.6)	35 (79.5)
Total number of samples	75	38	44

Table 5. Average salinity (% $\pm$ SD) of water where larvae were collected in Miyagi Prefecture, Japan during June to August 2011.

	June	July	August
An. sinensis-group	0.2	0.33 ± 0.05	0.06 ± 0.05
Cx. inatomii	—	0.25 ± 0.21	0.14 ± 0.19
Cx. tritaeniorhynchus	—	0.31 ± 0.12	0.2 ± 0.13
Cx. pipiens pallens	—	—	0.1 ± 0.1
Cx. orientalis	0	—	0
Cx. pseudovishnui	0	—	—

Differences in average salinity among mosquito species were not statistically significant ($F = 0.289$, $p = .755$ in July and $F = 2.024$, $p = 0.111$ in August).

graphic distribution of *Cx. inatomii* is also suggested from the results of mosquito surveys conducted by the Quarantine Station in Japan (Mizuta et al., 2012). Furthermore, the distribution of *Cx. inatomii* outside Japan was reported in Korea and China (Ree, 1998; Xu et al., 1993); therefore, *Cx. inatomii* has a wider geographic distribution than previously expected. Although the distribution of *Cx. inatomii* has not been reported in Miyagi Prefecture, the lack of collection records may be because of few mosquito studies conducted in the current study area in the past. Small populations of *Cx. inatomii* have probably been maintained in temporal ground pools appearing beside the seashore in Miyagi Prefecture, and they were the source populations of the outbreak observed in this study. The ground pools widely present in the Tsunami area provided good larval habitats for *Cx. inatomii* and resulted in the density increase and the expansion of spatial distribution, because *Cx. inatomii* larvae develop well in brackish water and adults reproduce autogenously (Katano et al., 2010).

As shown in Fig. 3, the spatial distribution of host-seeking *Cx. inatomii* was restricted to near the larval habitats. One of the species related to *Cx. inatomii* occurring in the Mediterranean area, *Cx. modestus* Ficalbi, has similar ecology to *Cx. inatomii* and its flight range is considered to be 500 to 700 m (Pradel et al., 2009; Cailly et al., 2011; Becker et al., 2010). Therefore, the flight range of *Cx. inatomii* seems to be the same as *Cx. modestus*, and a low chance of contact with humans is expected at present, because temporary dwellings are constructed in hilly areas far away from the larval habitats of *Cx. inatomii*. The blood meal of blood-fed *Cx. inatomii* in this study was identified as *R. norvegicus*, indicating contact with wild animals.

The reason for the high density of *Cx. pipiens pallens* in the Tsunami area is unclear at present. When the Tsunami destroyed the study area, adults of *Cx. pipiens pallens* were still in diapause, because the cumulative average temperature from the beginning of the year to 8 March 2011 in Sendai, Miyagi was 123.4, quite lower than the value suggested for the appearance of overwintered *Cx. pipiens pipiens* in the USA, 400 degree days (Bolling et al., 2007; Ciota et al., 2011). There are no available data on the appearance day of overwintered *Cx. pipien pallens* in Miyagi Prefecture. During the course of sweeping collections of overwintered *Cx. tritaeniorhynchus* in Tokyo (Tsuda and Kim, 2010), some overwintered females of *Cx. pipiens pallens* were found in the sweeping samples and were available for estimation of the appearance day of *Cx. pipiens pallens* in Tokyo as a reference. The first overwintered female of *Cx. pipiens pallens* was collected on 16 March in 2008, 7 March in 2009 and 5 March in 2010 and the cumulative average temperature from the beginning of the year was 502, 479.8 and 442.5 degree days, respectively. The cumulative degree days in Sendai, Miyagi Prefecture this year reached 400 degree days at the end of April; thus, it is likely that *Cx. pipiens pallens* were still in diapauses and overwintering when the Tsunami disaster occurred in Miyagi Prefecture on 11 March 2011. It is known that overwintering *Cx. pipiens pallens* can be found in caves and/or cellars (Hayashi et al., 1965; Shimogama and Takatsuki, 1967; Makiya and Sakurai, 1975); therefore, *Culex pipiens pallens* in these overwintering sites situated outside of the Tsunami area were able to escape the disaster and successfully overwintered and appeared at the end of April 2011. The drainage systems around human dwellings and irrigation cannels in rice field areas were destroyed by the earthquake and Tsunami, and various water bodies of different sizes formed on the ground. Some of these water bodies were good larval habitats for *Cx. pipiens pallens* and produced a large number of adults observed in July and August 2011, although we found *Cx. pipiens pallens* larvae in only 2% (3/157) of larval samples. Nearly all ground pools in the Tsunami area examined in this study contained brackish water and this result partly explains the small number of larvae of *Cx. pipiens pallens* collected at our study site.

A high occurrence of *Cx. tritaeniorhynchus* larvae in brackish ground pools was found in this study. The result indicates that *Cx. tritaeniorhynchus* larvae are capable of developing in brackish water. Since no studies have been carried out on the effects of the salinity of rearing water on the development and survival of larvae of *Cx. tritaeniorhynchus*, laboratory studies will be required to evaluate the after-effects of the Tsunami on the adult density of *Cx. tritaeniorhynchus* in the future.

The adult density of *Ae. albopictus* was significantly lower in the Tsunami area than in the No Tsumani area, indicating the destructive effects of the Tsunami on larval habitats of *Ae. albopictus*, which are located mainly beside human dwellings, such as in water catchments, earthenware pots, stone vases, used tires, bamboo stumps etc. These larval habitats were destroyed and flushed out by the sea wave; therefore, *Ae. albopictus* populations persisting in the No Tsunami area are considered to be the source population. The dispersal or flight ability of insects is an important ecological character determining their colonizing ability. The flight range of *Ae. albopictus* is usually short, less than 100 m (Hawley, 1988), indicating the low colonizing ability of *Ae. albopictus* in the disaster areas, and thus, only a small number of adult *Ae. albopictus* succeeded in immigrating into the disaster area during the summer season. Some of the immigrated *Ae. albopictus* will become source populations next year and density increase and expansion of the spatial distribution is expected because the potential larval habitats of *Ae. albopictus* are abundant in the disaster areas. Continuous adult mosquito surveillance in next season will be necessary to confirm the colonization and re-establishment of *Ae. albopictus* in disaster areas with human activities.

The habitat destruction of *Ae. vexans nipponii* was also suggested in the present study. Although only one female *Ae. vexans nipponii* was collected in this study, Kato et al. (1967) reported *Ae. vexans nipponii* as one of the dominant mosquitoes in rice field areas in Miyagi Prefecture in 1965. This species overwinters as eggs and the overwintering eggs are laid in soil; therefore, it is likely that eggs laid in rice fields were flushed out by the Tsunami and the density of the overwintered generation decreased greatly this year.

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