The distribution of the invasive shrimp *Neocaridina davidi* (Decapoda: Caridea: Atyidae) in relation to environmental parameters in a stream at Kunitachi, Tokyo, Japan

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**Abstract.**—The distribution of the invasive freshwater shrimp, *Neocaridina davidi*, in a spring-fed stream at Kunitachi, Tokyo, Japan, was investigated in relation to the stream’s environmental parameters. Compared to previous studies, the population density of *N. davidi* was found to be remarkably high (up to 315 ind./m²). The increase in population density is likely because the water temperature in the spring-fed stream remains 17–20°C in the winter, prolonging the period during which the shrimps can spawn. Generalized linear mixed models showed that adult shrimps were more abundant in areas with high emergent plant coverage. In contrast, juveniles were more abundant in areas with low current velocity. These results provide essential clues to the interspecific relationships between *N. davidi* and native spring organisms that inhabit the same physical environments.

**Key words:** Atyidae, invasive species, microhabitat, springs, Tokyo

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**Introduction**

Species of *Neocaridina* are landlocked freshwater shrimps widely distributed in East Asia (Cai, 1996). Used as fishing bait and ornamental species, they have spread across the world (Englund & Cai, 1999; Niwa, 2010; Klotz et al., 2013; Jabłońska et al., 2018; Weiperth et al., 2019). In Japan, several invasive species of *Neocaridina*, including *N. davidi*, were found in western Japan in the early 2000s (Niwa et al., 2005; Niwa, 2010; Fujita et al., 2011; Toyota & Seki, 2014). These species have recently spread to eastern Japan (Kanazawa, 2015; Nishida, 2016; Katayama et al., 2017; Mitsugi et al., 2017). The introduction and expansion of the invasive *Neocaridina* spp. in Japan could lead to interbreeding with, and the competitive exclusion of, native shrimps (Niwa, 2010; Nishida, 2016; Katayama et al., 2017). However, few studies have examined the interspecific relationships between the invasive *Neocaridina* spp. and the native organisms.

The recent invasion of *N. davidi* has been observed at Mama-shita springs (=Mama-shita Yusui) in Kunitachi, Tokyo, Japan, where this study was conducted. Mama-shita springs are critical components of Tokyo’s aqueous environment (Ueda et al., 2000; Takamura & Marui, 2014). The springs are home to several rare spring-dependent species, including Japanese eight-barbel loach *Lefua echigonia* and Amur minnow *Rhynchocypris lagowskii steindachneri* (Nishida et al., 2014). However, it is unclear how *Neocaridina* spp. affect the native organisms in the unique environment of a spring. For example, the habitat of *Neocaridina* spp., which can shed light on the interaction between the invasive and native population, is unknown.

In this study, I examined the distribution of *N. davidi*, a representative of the invasive *Neocaridina* spp., in the spring-fed stream in the Mama-shita Springs Park (=Mama-shita Yusui...
Koen) in Kunitachi, Tokyo, Japan, and investigated the effect of the stream’s physical environmental parameters on the distribution.

**Materials and Methods**

**Study site**

The study was conducted monthly in a spring-fed stream in the Mama-shita Springs Park (35°24′N, 139°25′E) at Kunitachi, Tokyo, Japan, from September to November 2019. The stream is approximately 300 m in length and joins the Fuchu waterway. In the stream, 12 study sections (Sec. 1–Sec. 12), each of a constant area of 2 m$^2$ in September and November and 4 m$^2$ in October, were set up (Fig. 1).

**Sampling shrimp**

Sampling was carried out at each study section by settling a small set net with a mesh size of 2 mm in the stream and blocking its flow. All samplings were performed by the same persons by cornering shrimps into the set net. The collected specimens were brought back to the laboratory, fixed in 10% neutral formalin, stained in Rose Bengal, and stored in 70% ethanol.

**Estimating the number of adult and juvenile shrimp**

The collected specimens were identified under a stereomicroscope. The adult and juvenile *Neocaridina davidi* were identified in each study section and counted. Identification of *N. davidi* was performed according to Toyota & Seki (2014). According to Mitsugi & Suzuki (2018), individuals with a carapace length of more than or less than 3 mm were defined as adults or juveniles, respectively.

The percentage of adult and juvenile *N. davidi* in each section was studied. One hundred individuals of *N. davidi* were randomly selected from the specimens of each study section to have their carapace length measured. For study sections with fewer than 100 sampled individuals, the carapace length of all the individuals was measured.

The number of adults and juveniles in each study section was calculated using the following equation;

\[ P_a = P_{all} \times \frac{P^*}{P_{all}}, \quad P_j = P_{all} \times \frac{P^*}{P_{all}}. \]

\[ P \] is the number of individuals in the entire study section, \( P^* \) is the number of individuals whose carapace length was measured, and the

Fig. 1. Map of the Mama-shita Springs Park (left) and the location of the study sections (right). Arrows indicate water flow direction.
indices all, a, and, j denote all individuals, adults, and juveniles, respectively.

Measuring physical environmental parameters

In September and October, surveys were carried out in all sections except for Sec. 4 and 9. In November, all 12 sections were surveyed. The emergent plant coverage, submerged plant coverage, current velocity, water depth, water temperature, and sediment score were examined in each study section using the following methods.

The respective coverage of the emergent plant, Japanese sweet flag *Acorus gramineus*, and submerged plant, Japanese bur-reed *Sagittaria japonica*, was calculated by dividing the plant community’s area in the study section by the area of the study section. The average current velocity of each study section was calculated by measuring the time required for a float to flow for 1 m at the center of the current three times. The average water depth of each study section was calculated by measuring the water depth at five randomly-selected points using a folded tape measure. Water temperature was measured using Quanta (HYDROLAB). Sediment was classified into four categories: large gravels (>64 mm), medium gravels (4 to 64 mm), fine gravel (2 to 4 mm), and fine sand (<2 mm); the area of coverage of each category was calculated. The sediment score $S$, which indicated the sediment roughness in each study section, was calculated by

$$S = \sum k_i C_i.$$  

The $k_i$ value is a constant given to the sediment category $i$ (large gravel: 4, medium gravel: 3, fine gravel: 2, fine sand: 1) and $C_i$ is the coverage of the sediment category $i$ (Dohi et al., 2006).

Statistical analysis

Two generalized linear mixed models (GLMM) were used to determine the effect of the stream’s physical environmental parameters on the population density of the adult and juvenile *N. davidi*. R 4.0.2 was used for all analyses (R Core Team, 2020).

The spatial autocorrelation variables were calculated to consider the effect of spatial proximity on the distribution of *N. davidi*. Moran’s eigenvector maps (MEM) were constructed using the relative neighborhood method, and the scores for each study section were calculated using the R package “adespatial” (Dray et al., 2020). MEM1 to MEM4 were used in the following analysis (Fig. 2).

A GLMM with a negative binomial distribution was constructed using the R package “MASS” to investigate the relationship between the density of the juvenile or adult *N. davidi* and the stream’s physical environmental parameters. The response variable was the number of juvenile or adult *N. davidi* in each study section. The explanatory variables were the scaled physical environmental parameters and MEM1 to MEM4, with an offset term accounting for the area of each study section. To avoid multicollinearity, Spearman’s correlation coefficients were calculated among all explana-
tory variables, and water depth and sediment scores, which have $\rho > 0.6$ with current velocity, were excluded from the explanatory variables. The model was selected using a multi-model inference approach. All the subsets of the models based on the global model were produced and ranked based on the Akaike’s information criterion (AIC). The best performing models with $\Delta$AIC $< 2$ were used to perform model averaging using the R package ”MuMIn” (Bartoń, 2020).

Results and Discussion

High Neocaridina davidi density

During the study, the invasive shrimp, *N. davidi*, and the crayfish, *Procambarus clarkii*, were collected (Table 1; Fig. 3(A)). *N. davidi* was dominant in all months, at 97.2% of the total decapod crustacean population in September, 96.9% in October, and 95.7% in November (Table 1). Adult *N. davidi* were found in all study sections. Juveniles were found in all study sections except in Sec. 1 in any month or Sec. 2 in September. The combined population density of adults and juveniles was highest in Sec. 8 in September at 315 ind./m$^2$ (Fig. 3(A)), which is higher than that of *N. denticulata* or invasive *N. davidi* in Japan in previous studies (Oh et al., 2003; Nishida, 2016).

In a study examining the field population dynamics of *Caridina cantonensis* (*N. serrata* in the original paper), there was a positive correlation between the proportion of ovigerous females amongst sexually mature females and water temperature (Dudgeon, 1985). Another study examined *N. davidi*, which invaded the Tomoe River in eastern Japan, and found that its estimated spawning season coincided with a period when the water temperature exceeded 15°C (Mitsugi & Suzuki, 2018). The water temperature at Mama-shita springs is around 17–20°C throughout the year (Nishida et al., 2014; Nishida, unpublished data), likely prolonging the spawning season of *N. davidi* and increasing the density of the shrimp. In fact, three and two ovigerous females were found in October and November, respectively, when no ovigerous females were found in Mitsugi & Suzuki (2018) (Table 1; Fig. 3(A)).

Physical environmental parameters affecting the distribution of adult and juvenile *N. davidi*

The physical environmental parameters of the stream were measured (Fig. 3(B)). The parameters such as the emergent plant coverage, submerged plant coverage, water depth, current velocity, and sediment score differed among the study sections. On the other hand, the water temperature was generally comparable in all study sections at about 19.7°C (Fig. 3(B)). Although spatial autocorrelation was detected, GLMM revealed that the population density of adult *N. davidi* was positively correlated with emergent plant coverage, whereas that of juveniles was negatively correlated with current velocity (Table 2). These results suggest that the adult shrimp density tended to be higher in sections with high emergent plant coverage. In contrast, juvenile shrimp density tended to be higher in sections with low current velocity. Some rare spring-fed fishes, such as *Lefua echigonia* and *Rhynchocypris lagowskii steindachneri*, are also thought to use the emergent plant cover. Since *N. davidi* was observed at high densities in such a physical environment, this species’ impact on native organisms needs to be investigated.

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Table 1. The total number of *Neocaridina davidi* and *Procambarus clarkii* collected in each month. In each species, the upper row shows the total number of individuals in all study sections, and the lower row shows the percentage to the total specimens. For *N. davidi*, OF indicates the number of ovigerous females.

|          | Sep.                  | Oct.                  | Nov.                  |
|----------|-----------------------|-----------------------|-----------------------|
| Neocaridina davidi | 2038 (OF: 12) | 1266 (OF: 3) | 3068 (OF: 2) |
|          | 97.2%                 | 96.9%                 | 95.7%                 |
| Procambarus clarkii | 58                   | 40                    | 137                   |
|          | 2.7%                  | 3.1%                  | 4.3%                  |

Fig. 3. (A) The distribution of adult (upper panels) and juvenile (lower panels) *Neocaridina davidi*. The number above the bar in the upper panels indicates the number of ovigerous females. (B) The physical environmental parameters in each study section in each month. Crosses indicate not collected or not measured.
Table 2. The results of model averaging of the selected general linear mixed model (GLMM) with the Akaike’s information criterion (ΔAIC<2).

| Explanatory variables              | Adults          |                  |                   | Juveniles |                  |                   |
|------------------------------------|-----------------|-----------------|-------------------|-----------|-----------------|-------------------|
|                                    | Estimate        | z                | Pr(>|z|)           | Estimate  | z                | Pr(>|z|)           |
| Emergent plant coverage            | 0.35            | 2.58 **          |                   | -0.06     | 0.29             |                   |
| Submerged plant coverage           | 0.25            | 1.72             |                   | 0.20      | 1.42             |                   |
| Current velocity                   | -0.04           | 0.23             |                   | -0.41     | 2.46 *           |                   |
| Temperature                        | -0.30           | 1.87             |                   | -0.10     | 0.63             |                   |
| MEM1                               | 0.44            | 2.47             | *                 | 0.93      | 4.57 ***         |                   |
| MEM2                               | -0.46           | 3.74             | ***               | -0.90     | 5.12 ***         |                   |
| MEM3                               | 0.04            | 0.27             | ***               | 0.36      | 2.59 **          |                   |
| MEM4                               | 0.53            | 3.53             | ***               | 0.58      | 3.49 ***         |                   |

*Pr(>|z|)<0.05, ** Pr(>|z|)<0.01, *** Pr(>|z|)<0.001.

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