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Toxicity evaluation of road dust in the runoff process using a benthic ostracod *Heterocypris incongruens*

Haruna Watanabe [a,*], Fumiyuki Nakajima [b], Ikuro Kasuga [a], Hiroaki Furumai [a]

[a] School of Engineering, The University of Tokyo, Hongo 7-3-1, Bunkyo-ku, Tokyo, 113-8656, Japan
[b] Environmental Science Center, The University of Tokyo, Hongo 7-3-1, Bunkyo-ku, Tokyo, 113-0033, Japan

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ABSTRACT

Road dust is considered an important source of sediment contamination; however, there are few studies on the toxicity of road dust on benthic organisms. This study evaluates the toxicity of road dust on the benthic ostracod, *Heterocypris incongruens*, through a 6-day direct exposure experiment. All six road dust samples collected in heavy traffic areas caused high mortality of the ostracod, whereas the road dust collected in a residential area did not show toxicity to the ostracod. After maintaining the mixture of road dust and water for 24 h, the toxicity of the road dust decreased significantly for three of the six samples in the heavy traffic areas, suggesting these road dust samples become less toxic in the surface runoff process. In addition, we conducted the same toxicity test on manipulated road dust using different solid/liquid ratios and holding times to evaluate the change in toxicity caused by the runoff process. Wet road dust that had been separated from a dust–water mixture after a holding time of 1 h or 24 h did not show lethal toxicity, while the water-soluble fraction of the mixture caused high mortality of the ostracod at a solid/liquid ratio of 1:2 and 1:4. However, after a 7-day holding time of the dust–water mixture, the wet road dust and the water-soluble fraction showed lethal toxicity to the ostracod. These results suggest that toxicants of road dust seemed to exist mainly in water soluble fraction eluted off by rain water; however, particle-bound fraction again showed lethal toxicity after 7 days of incubation.

1. Introduction

Contaminants have been introduced into sediments in urban waters through several routes (e.g., effluent discharge, ocean and lake disposal, nonpoint sources, contaminant spills, and airborne deposition) (Power and Chapman, 1992). Of these contaminants, road dust is one of the major nonpoint sources of pollution in urban areas. Its sources include pavements, paint materials, leakage of fuel, tire debris, brake-lining debris, exhaust particles, and vehicle components (Sator and Boyd, 1972). Previous studies have shown that road dust contains high concentrations of various toxic substances such as heavy metals, polycyclic aromatic hydrocarbons (PAHs), and perfluorinated surfactants (PFSs) (Takada et al., 1991; Lau and Stenstrom, 2005; Murakami et al., 2005, 2008; Murakami and Takada, 2008). Contaminated road dust is often flushed into the surface waters through road runoff and is considered an important source of sediment contamination. Faure et al. (2000) showed that road asphalt was a source of petroleum by-products pollution in river sediments. This was demonstrated by an analysis of polycyclic biomarkers and microscopic observation capable of identifying asphalt particles in river sediments. Thus, it is necessary to evaluate the toxicity of road dust to aquatic organisms as an important source of sediment contamination.

Most of the previous studies evaluated the toxicity of the water-soluble fraction of road dust, i.e. the road runoff water using bacteria and pelagic organisms such as green algae, cladoceran, and fish (Schiff et al., 2003; Greenstein et al., 2004; Kayhanian et al., 2008). Application of the toxicity identification evaluation (TIE) methods (US-EPA, 1991, 1996) using the purple sea urchin (*Strongylocentrotus purpuratus*) demonstrated that zinc was the major toxicant in the road runoff (Schiff et al., 2003; Greenstein et al., 2004). On the other hand, to evaluate the toxicity of the water-insoluble fraction of road dust, Ono et al. (1999) conducted the *umu*-test and the toxicity test using green algae, *Pseudokirchneriella subcapitata* and the crustacean, *Thamnocephalus platyurus,* on an organic solvent extract of road dust. The extract of particles in the road runoff and the road dust showed high genotoxicity related to nitrated polycyclic aromatic hydrocarbons (NO_2-PAHs) and also showed toxicity to the algae and the crustacean. However, the organic solvent extract of road dust may overestimate the bioavailability of organic compounds and exclude inorganic compounds as toxicants. Furthermore, there are few evaluations of the toxicity of road dust using benthic organisms. Maltby et al. (1995a,b) found that the discharge of road runoff had an adverse effect on the diversity and composition of macroinvertebrates

* Corresponding author. Tel.: +81 3 5841 6242; fax: +81 3 5841 6244.
E-mail addresses: h_watanabe@env.t.u-tokyo.ac.jp (H. Watanabe), nakajima@env.t.u-tokyo.ac.jp (F. Nakajima), kasuga@env.t.u-tokyo.ac.jp (I. Kasuga), furumai@env.t.u-tokyo.ac.jp (H. Furumai).

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in a small stream as the concentrations of heavy metals and PAHs in the sediment increased. They also conducted an amphipod toxicity test using *Gammarus pulex* on the organic solvent extract of sediments upstream and downstream of the runoff discharge. This showed an increase in the mortality of the amphipod in the extract downstream of the runoff discharge. However, as mentioned above, it is likely that the organic solvent extract of the sediment does not reflect the actual toxicity derived from the sediment. Therefore, whole-sediment toxicity tests in which benthic organisms are directly exposed to the sediment can be used to assess precisely the toxicity of the whole sediment (Hansen et al., 2007). Similarly, to assess the toxicity of whole road dust on benthic organisms in the aquatic environment, whole-sediment toxicity tests need to be performed.

It is also important to understand how the toxicity changes in the road dust in the runoff process. In the runoff process, road dust is washed off the road surface into drainage systems by rain water, sometimes retained in the drainage systems for a while, and finally transported to the receiving water. Runoff behavior of road dust and its associated pollutants depends on several factors including rainfall intensity, street surface characteristics, particle size (Sator and Boyd, 1972; Furumai et al., 2002) and structure of drainage systems. In addition, these factors can affect the change of chemical and physical characteristics of road dust during the runoff process, resulting in the change of the toxicity of road dust. If we find out the condition under which the road dust becomes toxic in the runoff process, we can develop effective countermeasures against the discharge of toxic road dust to the receiving water. Greenstein et al. (2004) showed that the toxicity of the runoff from a parking lot in the sea urchin fertilization was affected by antecedent dry weather period, rainfall intensity, and rainfall duration. However, the toxicity change of the road dust in the runoff process under different conditions has not been elucidated yet.

In this study, we evaluated the toxicity of road dust by direct exposure of a benthic ostracod, *Heterocypris incongruens*. Seven road dust samples were collected from different streets in Tokyo, and the toxicity was compared among sampling sites. In addition, to understand how the toxicity of road dust changes in the runoff process, we conducted the toxicity test on road dust under different conditions simulating the runoff process. The effect the amount of rainfall had in the washing-out process on the road was evaluated by decreasing the solid/liquid ratio of the road dust and water. Holding time in the drainage systems was also investigated by increasing the holding time of the road dust and water mixture before exposing the ostracod. We also conducted the toxicity test on the water phase and wet road dust. These were separated from the whole road dust by centrifugation to evaluate whether either the water phase or the solid phase caused the toxicity to the ostracod.

2. Materials and methods

2.1. Samples

Road dust samples were collected using a vacuum cleaner from the road gutter surface of six streets (Sts. 1, 2, 4–7) and from a highway parking area (St. 3) in Tokyo, Japan. Table 1 shows the sampling locations, dates, traffic and weather conditions, and the amount of road dust per surface area. The collected samples from Sts. 1–5 and 7 were sieved through a 2-mm mesh to remove large particles and stones. For the St. 6 sample, two road dust samples collected from Koishikawa Street and Shinobazu Street were sieved through a 2-mm mesh and mixed in a dry weight ratio of 1:1 (Murakami et al., 2008). The mixed road dust from St. 6 was sieved through a 106-μm mesh after three years of storage in a refrigerator at 5 °C. All samples were stored in a refrigerator at 5 °C until the toxicity test was performed.

2.2. Sample preparation

Road dust samples were prepared under eight different conditions as described in Table 2. The seven road dust samples were mixed with synthetic freshwater (US-EPA, 1993) in a 1:2 (v/v) ratio and stirred with a plastic spoon for about 1 min. The dust–water mixtures were kept still for 1 h or 24 h at 25 °C in darkness (corresponding to 'Holding time before exposure' in Table 2). A 1-h holding time was based on the protocol of the sediment toxicity test using the ostracod (Section 2.3). However, almost all standard sediment toxicity tests described in the US-EPA guidelines (US-EPA, 1994, 2000) require maintaining the mixture for 24 h before exposing the test organisms. After the holding time, the same volume of an algal suspension (Section 2.3) as synthetic freshwater was added as food for the ostracod 20 min before the exposure started. For the road dust from St. 6, the toxicity test was conducted with a dilution series to calculate LC50 (Median Lethal Concentration). The road dust was diluted with fine quartz (Merck KGaA, Cat No.: 107536) to the concentrations of 3.125%, 6.25%, 12.5%, 25%, 50%, and 100% (v/v). These samples were

Table 1
Sampling location and conditions.

Sampling site	Coordinate (WGS84)	Area type	Traffic density[a] (vehicles per day)	Sampling date	Antecedent dry weather period [b] (h)	Amount of road dust (g/m^2)
St. 1 (Kasuga Street)	N 35°70′ 75″ E 139°76′38″	Heavy traffic area	28,392	May 6, 2010	168	1.8
St. 2 (Keihin Highway No.1)	N 35°64′98″ E 13°975′35″	Heavy traffic area	59,461	May 17, 2010	138	1.5
St. 3 (Tatsumi PA No. 1)	N 35°64′34″ E 139°80′97″	Heavy traffic area	64,715	May 17, 2010	140	22.9
St. 4 (Hakusan Street)	N 35°70′81″ E 139°75′38″	Heavy traffic area	56,069	May 14, 2010	54	5.9
St. 5 (Kiyosumi Street)	N 35°69′10″ E 139°79′58″	Heavy traffic area	28,718	May 17, 2010	142	25.7
St. 6 [c] (Koishikawa Street)	N 35°43′07″ E 139°44′37″	Heavy traffic area	17,530	Nov 6, 2004	103	0.4
(Shinobazu Street)	N 35°43′07″ E 139°45′51″		21,607	Nov 5, 2004	125	0.4
St. 7 (Mitaka City)	N 35°41′14″ E 139°33′04″	Residential area	–[d]	June 25, 2010	44	158.8

[a] Traffic density at the closest available monitoring station to each sampling site was obtained from a public database (Japan Society of Traffic Engineers., 2006).
[b] Antecedent dry weather period was defined as the period when precipitation was lower than 0.5 mm at the Tokyo station. Data was obtained from a public database (Japan Meteorological Agency, 2010).
[c] Data of St. 6 sample was obtained from Murakami et al. (2008).
[d] "–": Data not available.