

Marine sediment conservation using benthic organisms

Katsutoshi ITO^{*1}, Mana ITO^{*1}, Takeshi HANO^{*1}, Toshimitsu ONDUKA^{*1}, Kazuhiko MOCHIDA^{*1},
Nobuaki SHONO^{*2}, and Ryuhei NAKAMURA^{*2}

Abstract: Estuaries and coastal zones used for fish aquaculture are often polluted by organic contaminants and anthropogenic chemicals. Bioremediation has been recognized as an efficient technology to clear environmental pollutants. The authors therefore studied environmental remediation using benthic organisms and have shown that some annelids are adaptable to polluted environments. For example, *Capitella* cf. *teleta*, a polychaete inhabiting the sediment beneath a fish farm, displayed high protease activity, whereas *Perinereis nuntia*, a polychaete inhabiting an estuary, displayed high cellulase activity. Additionally, the oligochaete *Thalassodrilides* cf. *briani* was found to survive highly hypoxic and sulfidic sediments contaminated with various pollutants and was shown to biotransform 1-nitronaphthalene, a toxic and carcinogenic chemical, into substances that are nontoxic to fish.

In another experiment in which these three benthic species were maintained in polluted sediments, the polychaetes *P. nuntia* and *C. cf. teleta* markedly increased redox potential (Eh) and decreased the level of acid volatile sulfides relative to the oligochaete *T. cf. briani*. Furthermore, the concentration of polycyclic aromatic hydrocarbons (PAHs) in the sediment with all three species was significantly lower than the initial level. *T. cf. briani*, especially, showed a marked ability to degrade the PAHs in the sediment. These results indicate that benthic organisms have species-specific remediation properties and ecological functions in organically polluted sediments.

We are also working on the development of a real-time measuring device for determining the Eh in the sediment under fish farms as Eh is a comprehensive parameter to monitor the degree of contamination of these sediments.

Key words: annelid, bioremediation, environmental pollutants, Polychaeta, *Oligochaeta*, polycyclic aromatic hydrocarbons

Introduction

Estuaries and coasts are crucial for the life histories of many aquatic organisms. The estuarine and coastal sediments accumulate organic matter from both marine and terrestrial sources (Jorcin, 2000; Hu *et al.*, 2009; Zhang *et al.*, 2009). In the sediments of coastal areas where aquaculture is conducted, considerable organic enrichment is caused by the input of large amounts of unconsumed fish food and fish feces.

This eutrophication causes anoxic conditions and an increase in sulfides (Pawar *et al.* 2002; Tanigawa *et al.*, 2007), leading to serious problems, such as algal blooms and the elimination of the benthic community and seagrass (Holmer *et al.*, 2008; Leon *et al.*, 2010). Polycyclic aromatic hydrocarbons (PAHs), which are products of the incomplete combustion of fossil fuels, contaminate estuarine and coastal sediments in areas associated with human activity (WHO, 1998). Therefore, remediation of organically polluted

2018年8月31日受理 (Accepted on August 31, 2018)

^{*1} National Research Institute of Fisheries and Environment of Inland Sea, Fisheries Research and Education Agency, 2-17-5 Maruishi, Hatsukaichi, Hiroshima 739-0452, Japan

^{*2} Biofunctional Catalyst Research Team, RIKEN Center for Sustainable Resource Science (CSRS), 2-1 Hirosawa, Wako, Saitama 351-0198, Japan
E-mail: katsuit@affrc.go.jp

sediment is necessary for maintaining ecological balance, biological diversity, and the sustainable development of aquaculture.

One pollutant of interest is 1-nitronaphthalene (INN), a nitrated polycyclic aromatic hydrocarbon (nitro-PAH) that is formed during the incomplete combustion of organic compounds (Nielsen, 1984). Such compounds are formed mainly by the reaction of PAHs with nitrogen oxides in polluted air (Atkinson and Arey, 1994). Nitro-PAH products are more toxic to many organisms than are the parent PAHs (Yaffe *et al.*, 2001). Yaffe *et al.* (2001), using an environmental model, reported that INN had the highest concentration potential among nitro-PAHs in the Los Angeles Basin in Southern California, USA.

Macrofauna, especially sediment-dwelling polychaete and oligochaete worms, are known to affect sediment characteristics biologically, chemically, and physically through feeding activity, bioturbation, ventilation, and irrigation, resulting in mineralization and organic degradation in the sediments (Banta *et al.*, 1999; Volkenborn *et al.*, 2007; Giere, 2006; Heilskov *et al.*, 2006; Quintana *et al.*, 2011). The activities of some polychaetes are closely related to the remediation of contaminated sediments and water (Licciano *et al.*, 2005; Palmer, 2010).

Organic matter in the environment is decomposed chemically by many enzymes (e.g., proteases, cellulase phosphatases, and carbohydrases). Some enzymes have been utilized to estimate organic matter decomposition in water and sediment in estuarine and coastal environments (Hiroki *et al.*, 2003; Arnosti *et al.*, 2009). The polychaete *Capitella teleta* (formerly *Capitella* sp. I; Blake *et al.*, 2009) has been reported to remediate organically contaminated sediment by enhancing the decomposition rate of the organic matter of the sediment under fish farms (Tsutsumi and Montani, 1993; Kinoshita *et al.*, 2008). Moreover, some benthic organisms, including *C. teleta* and *Nereis diversicolor*, can degrade oil, acyclic hydrocarbons, and the PAHs, such as fluoranthene and pyrene in sediment (Gilbert *et al.*, 1994; Grossi *et al.*, 2002; Madsen *et al.*, 1997; Christensen *et al.*, 2002), indicating that they can contribute to the remediation of sediments polluted by PAHs.

In this proceeding, we introduce research results targeting organic pollutants, chemical pollutants, and

the remediation of polluted sediment using annelids.

Materials and Methods

Animals

The benthic organisms used in this study (Fig.1) are as follows: the nereidid polychaete *Perinereis nuntia*, opportunistic polychaete *Capitella* cf. *teleta*, and oligochaete *Thalassodrilides* cf. *briani*. Nereidid worms, including *P. nuntia*, are common in intertidal and shallow marine waters and are widely distributed off the coasts of Asia and in the southern hemisphere (Wilson and Glasby, 1993; Muir and Hossain, 2014). *P. nuntia* is a large-sized species, very common in Japan, and is often found burrowing in sand under stones in estuaries and on sheltered beaches, sometimes in near-anaerobic conditions. This species is used as a model species for pollutants and has the potential to be used for wastewater treatment through its ability to reduce organic matter (Palmer, 2010). *C. cf. teleta* is frequently observed in the sediment under fish farms in Japan and is a small- to medium-sized species. *Capitella* species are opportunistic and can tolerate hypoxia and sediment toxicants (Gamenick *et al.*, 1998; Bach *et al.*, 2005; O'Brien and Keough, 2013).

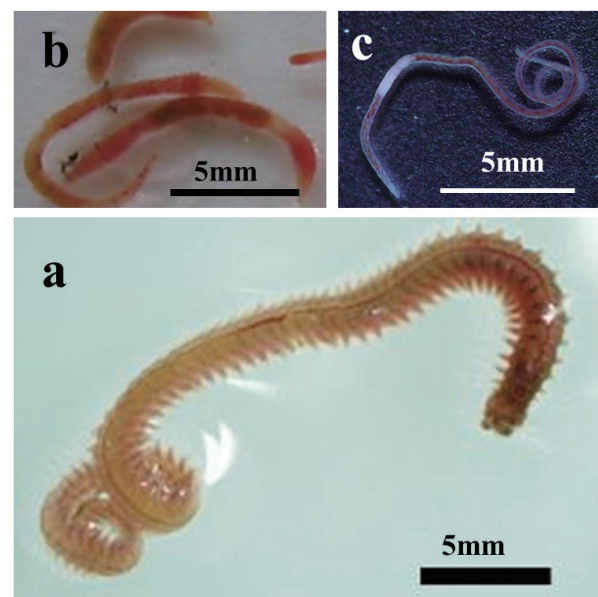


Fig. 1. Nereidid polychaete *Perinereis nuntia* (a), opportunistic polychaete *Capitella* cf. *teleta* (b), and oligochaete *Thalassodrilides* cf. *briani* (c).

T. cf. briani are distributed in the sediments of many areas including China, North America, and Western Australia (Erséus, 1990; Milligan, 1996).

Enzyme activities of the benthos

The whole body of each *P. nuntia* and *C. cf. teleta* worm, was homogenized in 10 mM sodium phosphate buffer (PBS; pH 7.5) using a pestle on ice. The homogenate was centrifuged for 30 min at 10,000 rpm at 4 °C, and then the supernatant was used for the determination of enzyme activities. Protease activity was measured using milk casein as a substrate according to the procedure of Kashiwagi (2004). Cellulase activity was measured at 37 °C for 24 h in 10 mM PBS (pH 7.5) with 0.8 % carboxymethyl cellulose as the substrate. Reducing sugars produced by the reaction were determined by the dinitrosalicylic reagent method (Miller 1959). Glucose was used as the standard.

Chemical pollutants catabolism tests with annelids

The residual INN in seawater following metabolism by both of the annelids was examined. Each replicate used 300 *T. cf. briani* individuals (total weight: 0.1 g), 5 *P. nuntia* individuals (average weight: 90 mg/individual; total weight 0.42 g). INN test solutions were made by adding the necessary amount of stock solution to pre-filtered (GFC filter, Whatman, Maidstone, UK) seawater. Animals were exposed to INN at 170 µg/L for 5 days in the dark at 20 °C in aquaria containing seawater and quartz sand. The residual INN concentration in seawater was analyzed by GC-MS at 0, 2, and 5 days.

Remediation of polluted sediment using annelids.

Artificial microcosms, consisting of clear glass columns (diameter 9.0 cm, height 12.2 cm) filled with 150 g of sediment (Sediment was collected from Hatsukaichi Marina of Hiroshima, Japan, an active harbor in which many fishing boats travel) and 300 mL seawater filtered through sand and activated carbon, were prepared. Approximately 150 mg biomass of one benthic worm species—*P. nuntia* (mean ± standard deviation) ($n = 5 \pm 0$), *C. cf. teleta* ($n = 59 \pm 5$), or *T. cf. briani* ($n = 160 \pm 6$)—was added to each of three microcosms. A fourth column, without benthic organisms, was prepared as a control. Three

replicate columns were prepared for each treatment.

The benthic worms were fed a commercial fish diet (N400; Kyowa Hakko, Tokyo, Japan) once every 3 days (5 % of total biomass per day; approximately 22.5 mg). The columns were closed and kept in an incubator at 20 °C for 50 days. For the initial sediment sample, a column was prepared as described above and kept overnight to allow the mud to settle. After removing the overlying water, the oxidation-reduction potential (ORP), acid volatile sulfides (AVS), loss on ignition (LOI measured as organic matter), and concentration of PAHs in the sediment were measured.

After 50 days, the overlying water was removed from all test microcosms, and the ORP (mV) of the sediment was measured at 2 cm depth using an ORP meter (D-55, Horiba, Kyoto, Japan). AVS was measured using a Hedorotech-S gas detection tube (GASTEC, Kanagawa, Japan). Benthic organisms were sorted from a subsample of the whole sediment sample. The living infauna were cleaned in seawater and their biomass measured; total column biomass was estimated from this measurement. The concentrations of 16 PAHs were measured in the whole sediment samples.

Results and Discussion

Enzyme activities of the benthos

The protease activity of *Capitella cf. teleta* (89.7 µg/mg) was about 10 times those of *P. nuntia brevicirris* (8.0 µg/mg). High cellulase (endo-β-1,4-glucanase) activity was detected in *P. nuntia brevicirris* (3.2 µg/mg), whereas the activity was scarcely detected in *Capitella cf. teleta*. The high protease activity of *Capitella cf. teleta* enabled it to survive in the sediment under a fish farm, where it degrades organic matter. In contrast, the high cellulase activity of the estuary-dwelling *P. nuntia brevicirris* allowed it to degrade organic matter originating from terrestrial areas.

Chemical pollutants catabolism tests with annelids

In the INN metabolism tests with annelids, the INN concentration in seawater did not decrease significantly from the initial concentration of 170 µg/L without animals present. However, in all animal treatments, the concentrations in the seawater decreased significantly (*T. cf. briani*: 23.3 %; *P. nuntia*:

32.6 %) after 2 days. After 5 days, the concentrations in the seawater were $3.0 \pm 0.2 \mu\text{g/L}$ in the *T. cf. briani* tank, and $14.3 \pm 2.1 \mu\text{g/L}$ in the *P. nuntia* tank. The ability of the *T. cf. briani* to biotransform INN was significantly greater than that of the *P. nuntia* ($p < 0.01$). Furthermore, *T. cf. briani* can survive in highly hypoxic and sulfidic sediments contaminated with various pollutants, and have been shown to biotransform INN, a toxic and carcinogenic chemical, into substances that are nontoxic to fish.

Remediation of polluted sediment using annelids

The annelid's effects on physicochemical properties, such as organic matter (LOI), Eh, AVS, and the degradation of PAHs, were assessed. Eh levels were significantly higher, and AVS levels lower, in the sediments of the polychaetes *P. nuntia* and *C. cf. teleta* than they were in those of the oligochaete *T. cf. briani* or the control (without benthic organisms). Total PAH concentration significantly decreased from the initial level in all three groups; *T. cf. briani* displayed a marked ability to reduce PAHs in sediment. These results indicate that benthic organisms have species-specific remediation properties and ecological functions in organically polluted sediments.

Real-time device for measuring Eh in sediment

In our group, sediment remediation tests using benthos were carried out in the field, such as at fish farms. Periodic surveys conducted by sediment sampling and diving were necessary to verify the effects of the annelids. The Eh is a comprehensive parameter, which can be used to monitor the degree of contamination of the sediment under a fish farm. Currently, we are working on the development of a real-time device for measuring Eh in the sediment under fish farms.

In fact, we are operating a real-time measurement in a 3-ton water tank and confirmed that the processes by which the organic matter in the sediments was being removed by remediation with annelids were successfully monitored, demonstrating that it is possible to measure redox potential in real time even in the field. In future, in the case of a fish farm where organic pollution has advanced, we plan to use an appropriate benthic species to restore the bottom

sediment. To observe the progress of the remediation, we plan to use real-time measuring to trace the Eh. If sediment pollution progresses, we will implement countermeasures, such as increasing the number of benthic organisms or decreasing the feed volume. This allows not only for evaluating the healthy conditions of benthic systems, but even facilitating the remediation potential of benthic organisms to improve the toluene of sediment ecosystems against over feeding on fish farms.

Acknowledgments

This work was supported by JSPS KAKENHI Grant Numbers 17K20075, 15K16144, 22710083, and 26712017.

References

- Arnosti C., Ziervogel K., Ocampo L., and Ghobrial S., 2009: Enzyme activities in the water column and in shallow permeable sediments from the northeastern Gulf of Mexico. *Estuar. Coast Shelf Sci.*, **84**, 202-208.
- Atkinson R. and Arey J., 1994: Atmospheric chemistry of gas-phase polycyclic aromatic hydrocarbons: formation of atmospheric mutagens. *Environ. Health Perspect.*, **102**, 117-126.
- Bach L., Palmqvist A., Rasmussen L. J., and Forbes V. E., 2005: Differences in PAH tolerance between *Capitella* species: Underlying biochemical mechanisms. *Aquat. Toxicol.*, **74**, 307-319.
- Banta G. T., Holmer M., Jensen M. H., and Kristensen E., 1999: Effects of two polychaete worms, *Nereis diversicolor* and *Arenicola marina*, on aerobic and anaerobic decomposition in a sandy marine sediment. *Aquat. Microb. Ecol.*, **19**, 189-204.
- Blake J. A., Grassle J. P., and Eckelbarger K. J., 2009: *Capitella teleta*, a new species designation for the opportunistic and experimental *Capitella* sp. I, with a review of the literature for confirmed records. *Zoosymposia*, **2**, 25-53.
- Christensen M., Andersen O., and Banta G. T., 2002: Metabolism of pyrene by the polychaetes *Nereis diversicolor* and *Arenicola marina*. *Aquat. Toxicol.*, **58**, 15-25.
- Erséus C., 1990: The marine Tubificidae (Oligochaeta)

- of the barrier reef ecosystems at Carrie Bow Cay, Belize, and other parts of the Caribbean Sea, with descriptions of twenty-seven new species and revision of *Heterodrilus*, *Thalassodrilides* and *Smithsonidrilus*. *Zool. Scripta.*, **19**, 243-303.
- Gamenick I., Vismann B., Grieshaber M. K., and Giere O., 1998: Ecophysiological differentiation of *Capitella capitata* (Polychaeta). Sibling species from different sulfidic habitats. *Mar. Ecol. Prog. Ser.*, **175**, 155-166.
- Giere O., 2006: Ecology and biology of marine Oligochaeta - An inventory rather than another review. *Hydrobiologia*, **564**, 103-116.
- Gilbert F., Rivet L., and Bertrand J. C., 1994: The in vitro influence of the burrowing polychaete *Nereis diversicolor* on the fate of petroleum hydrocarbons in marine sediments. *Chemosphere*, **29**, 1-12.
- Grossi V., Massias D., Stora G., and Bertrand J. C., 2002: Burial, exportation and degradation of acyclic petroleum hydrocarbons following a simulated oil spill in bioturbated Mediterranean coastal sediments. *Chemosphere*, **48**, 947-954.
- Heilskov A. C., Alperin M., and Holmer M., 2006: Benthic fauna bio-irrigation effects on nutrient regeneration in fish farm sediments. *J. Exp. Mar. Bio. Ecol.*, **339**, 204-225.
- Hiroki M., Yabe T., Nohara S., Utagawa H., Satake K., Koga T., Ueno R., Kawachi M., and Watanabe M., 2003: Evaluation of an organic matter decomposing function from hydrolytic enzyme activities of tidal flat sediments in Japan. *Jap. J. Limnol.*, **64**, 113-120. (in Japanese with English abstract).
- Holmer, M., Argyrou M., Dalsgaard T., Danovaro R., Diaz-Almela E., Duarte C. M., Frederiksen M., Grau A., Karakassis I., Marbà N., Mirto S., Pérez M., Pusceddu A., and Tsapakis M., 2008: Effects of fish farm waste on *Posidonia oceanica* meadows: Synthesis and provision of monitoring and management tools. *Mar. Pollut. Bull.*, **56**, 1618-1629.
- Hu L., Guo Z., Feng J., Yang Z., and Fang M., 2009: Distributions and sources of bulk organic matter and aliphatic hydrocarbons in surface sediments of the Bohai Sea. *China. Mar. Chem.*, **113**, 197-211.
- Jorcin A., 2000: Physical and chemical characteristics of the sediment in the estuarine region of Canan' eia (SP), Brazil. *Estuaries*, **431**, 59-67.
- Kashiwagi Y., 2004: Enzymes of filamentous fungi in fermentation. MAFF Microorganism Genetic Resources Manual No. 16, 1-16.
- Kinoshita K., Tamaki S., Yoshioka M., Srithonguthai S., Kunihiro T., Hama D., Ohwada K., and Tsutsumi H., 2008: Bioremediation of organically enriched sediment deposited below fish farms with artificially mass-cultured colonies of a deposit-feeding polychaete *Capitella* sp. I. *Fish. Sci.*, **74**, 77-87.
- Leon S. D., Mae S., Diego-McGlone S., Lourdes M., and Reichardt W., 2010: Impact of polychaete infauna on enzymatic protein degradation in marine sediments affected by intensive milkfish farming. *Aquacult. Res.*, **41**, 11.
- Licciano M., Stabili L., and Giangrande A., 2005: Clearance rates of *Sabella spallanzanii* and *Branchiomma luctuosum* (Annelida: Polychaeta) on a pure culture of *Vibrio alginolyticus*. *Water Res.*, **39**, 4375-4384.
- Madsen S. D., Forbes T. L., and Forbes V. E., 1997: Particle mixing by the polychaete *Capitella* species 1: Coupling fate and effect of a particle-bound organic contaminant (fluoranthene) in a marine sediment. *Mar. Ecol. Prog. Ser.*, **147**, 129-142.
- Miller G. L. 1959. Use of dinitrosalicylic acid reagent for determination of reducing sugar. *Anal Chem*, **31**, 426-428.
- Milligan M., 1996: Identification manual for the aquatic Oligochaeta of Florida. Vol. II. Estuarine and nearshore marine oligochaetes. Florida Department of environmental Protection, Tallahassee, Florida, 239pp.
- Muir A. I. and Hossain M. M. M., 2014: The intertidal polychaete (Annelida) fauna of the Sitakunda coast (Chittagong, Bangladesh), with notes on the Capitellidae, Glyceridae, Lumbrineridae, Nephtyidae, Nereididae and Phyllocidae of the "Northern Bay of Bengal Ecoregion". *Zookeys*, **27**, 1-27.
- Nielsen T., 1984: Reactivity of polycyclic aromatic hydrocarbons towards nitrating species. *Environ. Sci. Technol.*, **18**, 157-163.
- O'Brien A. L. and Keough M. J., 2013: Detecting

- benthic community responses to pollution in estuaries: A field mesocosm approach. *Environ. Pollut.*, **175**, 45-55.
- Palmer P. J., 2010: Polychaete-assisted sand filters. *Aquaculture*, **306**, 369-377.
- Pawar V., Matsuda O., and Fujisaki N., 2002: Relationship between feed input and sediment quality of the fish cage farms. *Fish. Sci.*, **68**, 894-903.
- Quintana C. O., Hansen T., Delefosse M., Banta G., and Kristensen E., 2011: Burrow ventilation and associated porewater irrigation by the polychaete *Marenzelleria viridis*. *J. Exp. Mar. Bio. Ecol.*, **397**, 179-187.
- Tanigawa T., Tamashita A., and Koizumi Y., 2007: Effect of effluents from a new fish farming site on the benthic environment. *Bull. Fish. Res. Agen.*, **19**, 69-77.
- Tsutsumi H. and Montani S., 1993: Utilization of biological activities of capitellid polychaete for treatment of "hedoro" (organically enriched sediment) deposited on the marine bottom below fish net pen culture. *Nippon Suisan Gakkaishi*, **59**, 1343-1347.
- Volkenborn N., Hedtkamp S. I. C., van Beusekom J. E. E., and Reise K., 2007: Effects of bioturbation and bioirrigation by lugworms (*Arenicola marina*) on physical and chemical sediment properties and implications for intertidal habitat succession. *Estuar. Coast. Shelf Sci.*, **74**, 331-343.
- WHO, 1998: Selected non-heterocyclic polycyclic aromatic hydrocarbons. Environmental Health Criteria 202. World Health Organization, Geneva.
- Wilson R. S. and Glasby C. J., 1993: A revision of the *Perinereis nuntia* species group (Polychaeta: Nereididae). *Rec. Aust. Museum*, **45**, 253-277.
- Yaffe D., Cohen Y., Arey J., and Grosovsky A. J., 2001: Multimedia analysis of PAHs and nitro-PAH daughter products in the Los Angeles Basin. *Risk Anal.*, **21**, 275-294.
- Zhang L., Yin K., Wang L., Chen F., Zhang D., and Yang Y., 2009: The sources and accumulation rate of sedimentary organic matter in the Pearl River Estuary and adjacent coastal area, Southern China. *Estuar. Coast. Shelf Sci.*, **85**, 190-196.

Annotated bibliography

- (1) Ito K., Nozaki M., Ohta T., Miura C., Tozawa Y., and Miura T., 2011: Differences of two polychaete species reflected in enzyme activities. *Mar. Biol.*, **158**(6), 1211-1221.

Polychaetes constitute most of the benthic macroinvertebrates in estuarine and coastal environments. We investigated the utilization of organic matter in two polychaete species, *Capitella* sp. I and *Perinereis nuntia brevicirris*, living in different coastal habitats. The protease activity of *Capitella* sp. I (89.7 $\mu\text{g}/\text{mg}$) was about 10 times that of *P. nuntia brevicirris* (8.0 $\mu\text{g}/\text{mg}$). High cellulase (endo- β -1,4-glucanase) activity was detected in *P. nuntia brevicirris* (3.2 $\mu\text{g}/\text{mg}$), whereas scarcely any was detected in *Capitella* sp. I. We isolated cDNA clones of protease mRNA from *Capitella* sp. I and of cellulase mRNA from *P. nuntia brevicirris*. The high protease activity of *Capitella* sp. I enabled it to survive in the sediment under a fish farm, where it degrades organic matter. In contrast, the high cellulase activity of the estuary-dwelling *P. nuntia brevicirris* allowed it to degrade organic matter originating from terrestrial areas.

- (2) Ito K., Ito M., Onduka T., Ohta K., Torii T., Hano T., Mochida K., Ohkubo N., Miura T., and Fujii K., 2016: Differences in the ability of two marine annelid species, *Thalassodrilides* sp. and *Perinereis nuntia*, to detoxify 1-nitronaphthalene. *Chemosphere*, **151**, 339-344.

Bioremediation is a promising method for remediating environmentally polluted water. We investigated the abilities of two benthic annelid species to biotransform 1-nitronaphthalene, a nitrated polycyclic aromatic hydrocarbon. We used an oligochaete, *Thalassodrilides* sp. (Naididae), collected from the sediment beneath a fish farm and a polychaete, *Perinereis nuntia*, which was obtained from a commercial source. Populations of both organisms were exposed to 1400 $\mu\text{g}/\text{L}$ of 1-nitronaphthalene in seawater for 3 days in the dark at 20 °C. The concentration of the pollutant decreased to 12 $\mu\text{g}/\text{L}$ in the seawater containing the *Thalassodrilides* sp. and to 560 $\mu\text{g}/\text{L}$ in the seawater containing *P. nuntia*. The 1-nitronaphthalene concentration in the

bodies of the animals increased from 12 to 94 $\mu\text{g}/\text{kg}$ in *Thalassodrilides* sp. and from 0.90 $\mu\text{g}/\text{kg}$ to 38,000 $\mu\text{g}/\text{kg}$ in *P. nuntia*. After 3 days, 99 % and 40 % of the 1-nitronaphthalene had been biotransformed in the *Thalassodrilides* sp. and *P. nuntia* experimental groups, respectively. We then tested the acute toxicity of residual 1-nitronaphthalene from the same water using mummichog (fish) larvae. After the larvae had been exposed for 96 h, the percentage of apparently unaffected larvae remaining was 83.3 % in *Thalassodrilides* sp. group but only 16.7 % in the *P. nuntia* group. Clearly, of the two species we studied, *Thalassodrilides* sp. had a superior ability to convert 1-nitronaphthalene into substances that were nontoxic to mummichog larvae. Therefore, we recommend the use of this species for bioremediation of chemically polluted sediments.

(3) Ito M., Ito K., Ohta K., Hano T., Onduka T., Mochida K., and Fujii K., 2016: Evaluation of bioremediation potential of three benthic annelids in organically polluted marine sediment. *Chemosphere*, **163**, 392-399.

This study aimed to evaluate the possible remedial effects of three marine benthic annelids on organically polluted sediments from the waters of Hatsukaichi Marina, Hiroshima, Japan. Two polychaetes, *Perinereis nuntia* and *Capitella* cf. *teleta*, and an oligochaete, *Thalassodrilides* sp., were incubated in sediments for 50 days. Their effects on physicochemical properties, such as organic matter (loss on ignition), redox potential (Eh), acid volatile sulfides (AVS), and degradation of polycyclic aromatic hydrocarbons (PAHs), were assessed. The polychaetes *P. nuntia* and *C. cf. teleta* significantly increased Eh level and decreased AVS level compared with the oligochaete *Thalassodrilides* sp. and control (without benthic organisms). Total PAH concentration significantly decreased from the initial level in all three groups; *Thalassodrilides* sp. had a marked ability to reduce PAHs in sediment. These results indicate that benthic organisms have species-specific remediation properties and ecological functions in organically polluted sediments.