EnvironmentAsia
The international journal published by the Thai Society of Higher Education Institutes on Environment

Available online at www.tshe.org/ea/index.html
EnvironmentAsia 9(2) (2016) 48-54
DOI 10.14456/ea.2016.7

# Qualitative Assessment and Management of Microplastics in Asian Green Mussels (*Perna viridis*) Cultured in Bacoor Bay, Cavite, Phillipines

Cristian Ryan Argamino and Jose Isagani B. Janairo

*Biology Department, De La Salle University, 2401 Taft Avenue, Manila 0922 Philippines*

**Abstract**

Microplastics (> 5 mm) have gained popularity in research and the public eye in recent years. This is due to the fact that they contain persistent organic pollutants (POPs) which pose potential risks to the environment and human health. Bivalves, which are filter feeders, are considered to be good indicators of marine pollution. In this preliminary study, Asian green mussel (*Perna viridis*), an example of edible bivalve, cultured in Bacoor Bay, Cavite, Philippines was subjected to qualitative analysis to determine the presence of microplastics. Through microscopic analysis, microplastics were found present in the acid-digested mussel soft tissue. A management program is suggested for policy makers and stakeholders to reduce the negative impact of microplastic pollution to both humans and the marine environment.

***Keywords:*** microplastics; qualitative assessment; *Perna viridis*; marine pollution; environmental management

## 1. Introduction

Plastics are popular due to several properties such as weight, strength and cost. The rise of consumption over the past few decades has been a huge challenge in terms of controlling environmental pollution. The increase in the number of plastics amassed in the environment is mainly caused by their inert property (slow degradation rate) and usage (improper disposal of plastic waste) (Nor and Obbard, 2014). Despite their benefits, the contribution of plastic to environmental degradation has been significant in causing damage to marine organisms. Previous studies have shown that plastics (i.e. microplastics) are ingested at different trophic levels which can potentially cause bioaccumulation. This phenomenon can affect biological processes of marine organisms. In addition, food safety and human health are also at risk, if these affected organisms are consumed (Vandermeersch *et al*., 2015).

It has been estimated that up to about 12.7 million tons of plastic still ended up in the ocean in 2010 (Jambeck *et al*., 2015; Andrady and Neal, 2009; Van Cauwenberghe *et al*., 2015). The Philippines was ranked No. 3 by mass of mismanaged plastic waste following China and Indonesia. In 2010, plastic marine debris in the Philippines ranged from 0.28-0.75 MMT (Million Metric Tons) per year (Jambeck *et al*., 2015). Marine plastic debris primarily come from land-based sources (80%) through leakage while only marine aquaculture and fisheries only contribute 20% of these pollutants (Ocean Conservancy, 2015). In recent years, the emergence of a form plastic has been studied for its role in pollution - microplastics. Although there has been no universally accepted definition at the moment (Van Cauwenberghe *et al*., 2015), individual and group researches have contributed their own description of microplastics. In 2004, microplastic size was defined at around 20 microns (Thompson *et al*., 2004). In 2009, the size adapted was <5 mm. This characterization of microplastic size was also accepted by the Joint Group of Experts on the Scientific Aspects of Marine Environmental Protection (GESAMP) (Nor and Obbard, 2014). Other researchers have presented microplastics as particles with <1 mm size which is argued to be more instinctive since 'micro' refers to the micrometer range (Van Cauwenberghe *et al*., 2015). As a marine pollutant, microplastics exist in two forms - primary and secondary. Primary microplastics, directly move into water bodies through ground runoff. These are mainly composed of virgin plastic pellets, scrubbers, and microbeads. These plastic forms are abundant in cosmetic products like exfoliants as well as industrial abrasives (Andrady, 2011). On the other hand, secondary microplastics are degradation products of larger plastics (mesoplastic/macroplastic) which are already in the ocean or seashore. Microplastics in this form are produced via several pathways such as mechanical, photo (oxidative) and/ or biological degradation (Masura *et al*., 2015). Majority of microplastics in the marine environment are secondary microplastics formed through the weathering of plastic in the seashore. Plastics on beaches have a faster degradation rate due to the higher temperature (of sand) as compared to both plastics deep in the ocean or floating on the water surface (Andrady, 2011).

Mussels, locally known as *tahong*, are generally defined as mollusk bivalves which grow in the wild or through aquaculture. These suspension feeders are considered to be one of the best biological indicators of marine pollution because of various characteristics including their geographical distribution. Their tissues accumulate pollutants due to their feeding mechanism and their tendency to stay attached to surfaces which make them relatively inactive in terms of mobility thus rendering them as good pollution gauges. They can also be easily sampled and thus, allow for frequent experiments and monitoring (Vasanthi *et al*., 2012; Figueiras *et al*., 2002; Chase *et al*., 2001). The species of mussel used in this experiment is *Perna viridis* (*P.vidiridis*). *P. viridis* is a large species of mussel ranging from 8-16 cm (Rajagopal *et al*., 2006). Their habitat includes hard surfaces (rocks, pilings and floating docks, as well as sandy or muddy bottoms about a foot below the low tide mark) (McGuire and Stevely, 2009). Their diet includes microscopic phytoplankton, zooplankton and suspended organic detritus in the water (Rajagopal *et al*., 2006). In the Philippines, *P. viridis* or *tahong* is considered as one of the seven major aquaculture species (Food and Agricultural Organization, 2006). Aquaculture of tahong can be traced back to 1955 which was initiated by the Philippine Bureau of Fisheries and Aquatic Resources in Binakayan, Cavite (Yap, 1999). Although it is a source of food locally, discretion is advised for consumption due to their tendency for bioaccumulation of toxins substances detrimental to human health (McGuire and Stevely, 2009). Thus, determining the presence of microplastics in these marine species will not only be instrumental to profiling marine pollution but will also serve as a tool in improving guidelines for food safety.

In this study, we present our findings regarding the presence of microplastics in cultured bivalves available for human consumption. These particles were positively identified through microscopic analysis and other qualitative criteria. In addition, management procedures according to best practices were also discussed which will aid policy makers in reducing the risk posed by these pollutants to the environment and human health.


Figure 1. Map of Bacoor Bay (Google Maps)

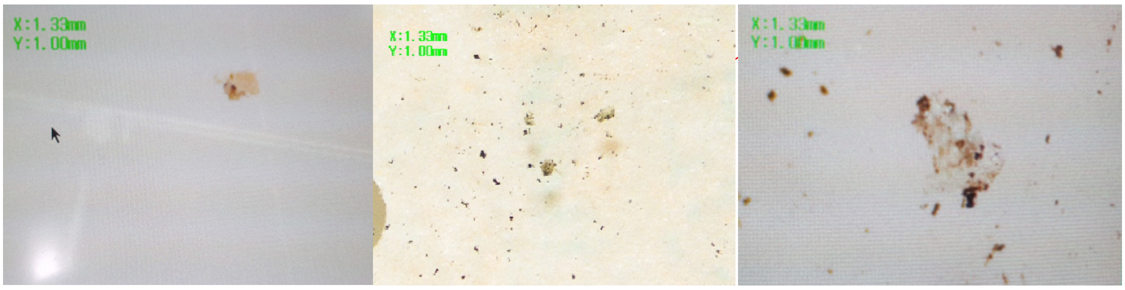

Figure 2. Semi-transparent microplastics

## 2. Materials and Methods

Bacoor Bay-cultured samples (*P.viridis*) were obtained from Sineguelasan Seafood Terminal in Bacoor, Cavite, Philippines. The mussel samples were harvested on January 2016. Fig. 1 shows a map of Metro Manila and its adjacent provinces. The drop pin marks the location of Bacoor Bay where the samples were cultured and harvested. All glassware were thoroughly cleaned and rinsed with filtered deionized water (Elga PURELAB Flex) to avoid contamination dilution (Van Cauwenberghe and Janssen, 2014).

Wet digestion using acid was used to extract the microplastics for the samples (Vandermeersch *et al*., 2015). Twenty milliliters (20 mL) of 70% $HNO_3$ (UNIVAR) were added to three mussels in an Erlenmeyer flask. Five replicates were prepared. Mussel tissues in acid were left in in the hood for 40 hours to achieve optimum digestion. The samples were then heated until boiling for 15-20 minutes using a hot plate to evaporate the acid and dry the sample. Twenty milliliters of warm deionized water (~80 degrees Celsius) were added to each vessel for dilution (Van Cauwenberghe and Janssen, 2014). Digested samples were then subjected to vacuum filtration using a Buchner funnel and Whatman Filter Paper Grade 1 (11-micron pore size). Filters were dried for 2 hours in an oven at 40 degrees Celsius. Dried filters were analyzed for the presence of microplastics using a Nikon SMZ 745T Stereomicroscope (Van Cauwenberghe and Janssen, 2014; Song *et al*., 2015).

## 3. Results and Discussion

Previous studies confirmed the presence of microplastics from marine samples using different methods. Raman Spectroscopy (Van Cauwenberghe and Janssen, 2014), Fourier Transform Infrared Spectroscopy coupled with either a Microscope or an Attenuated Total Reflectance attachment (Nor and Obbard, 2014), as well as the Fluorescence Microscope (Noren, 2007) were utilized by various studies. The absence of these instruments should not be a hindrance in the analysis of this important class of pollutants. Thus, the following criteria were used in determining the presence of microplastics (Noren, 2007):

a) No cellular or organic structures are visible in the plastic particle/fibre.

b) Clear and homogeneously colored particles (blue, red, black and yellow)

c) If the particle transparent or whitish, it shall be examined with extra care in a microscope under high magnification.

Figs. 2 and 3 show examples of microplastics observed in each of the five samples.

Microplastics observed in the samples were all found to be < 1mm. Transparent whitish and reddish particles measuring at around 10 to 30 microns (0.1 to 0.3 mm) were observed in Samples 1, 2, and 5. On the other hand, blue fibers (~0.5 mm in length) were observed in Sample 3 and 4. These observations qualify in the abovementioned criteria (Noren, 2007)

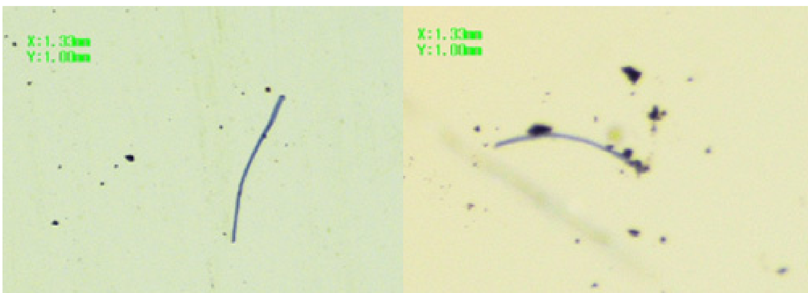

Figure 3. Fiber microplastics

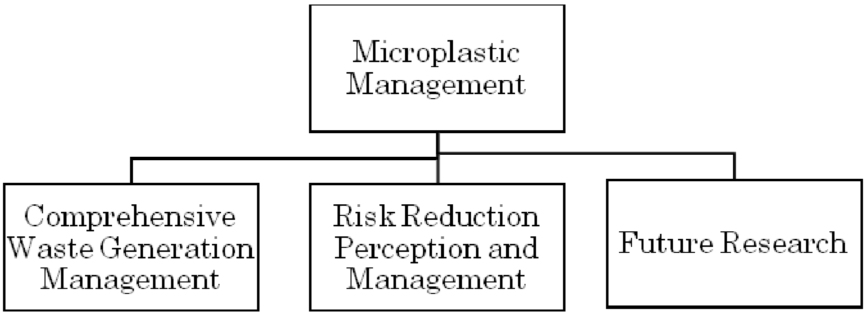


Figure 4. Microplastic Management

therefore, confirming the presence of microplastics in P. viridis. With the confirmation of the presence of microplastics in this preliminary study, further research on microplastics can now be focused on their quantitation and characterization using analytical tools such as the Fourier Transform Infrared Spectroscopy Microscope and Raman Spectroscopy. Microplastic presence needs to be addressed by policy makers and stakeholders. A suggested management program is summarized in Fig. 4 (GESAMP, 2015).

The first dimension of the program is a Comprehensive Waste Generation Management (CWGM) (Fig. 5) which can have a big impact in decreasing the amount of plastic in oceans. According to Jambeck *et al*. (2015), a 50% decrease in mismanaged waste from the Top 20 contributors (which includes the Philippines) will result to 41% mass decrease of mismanaged plastic waste by 2025. CWGM involves identification of hotspots or potential sources of the pollutant. In this case, microplastics ingested by bivalves can be attributed to sources such as urban inputs, fisheries, and aquaculture. Another aspect of CWGM is life cycle assessment which includes value-chain models and the popular 3R method (reduce, reuse, recycle). Life cycle assessment of plastics particularly the product disposal chain is an important task of management. This assessment will lead to a targeted approach which are prerequisites of an efficient and cost-effective method of reducing microplastic impact (Vegter *et al*., 2014). The 3Rs in particular should be targeted toward reducing marine debris by giving economic incentives to stakeholders. The last aspect of this program is the reduction of microplastic input (GESAMP, 2015). This can be implemented through policies which ban or regulate primary microplastics. The United States of America recently enacted a law "prohibiting the manufacture and introduction or delivery for introduction into interstate commerce of rinse off cosmetics containing intentionally-added plastic microbeads" (Microbead-Free Waters Act, 2015). Microbeads in cosmetic products are usually made up of polyethylene. Policies must also drive the reduction and prevention of secondary microplastics (formed from degradation) in entering the marine environment (Dippo, 2012). Research can also be driven towards the reduction of microplastic input. Plastics in marine environment are usually in the form of packaging materials. Further studies on alternative packaging materials either by significantly reducing the amount plastic used or redesigning and formulating alternative materials which can be easily and safely degraded to non-threating products (for both human and marine life) will decrease marine litter output. Physical screening and waste treatment technologies are also mitigation options to prevent microplastics from entering not only the marine environment but also groundwater which can pose risks to terrestrial animals and humans (Vegter *et al*., 2014). Fig. 5. shows Comprehensive Waste Management Program for Microplastics.

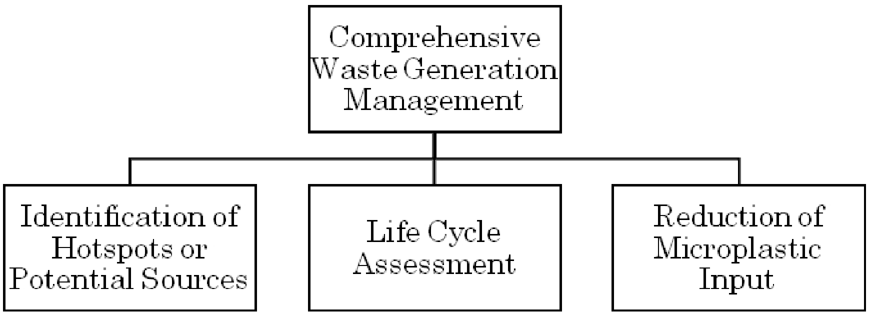


Figure 5. Comprehensive waste generation management plan

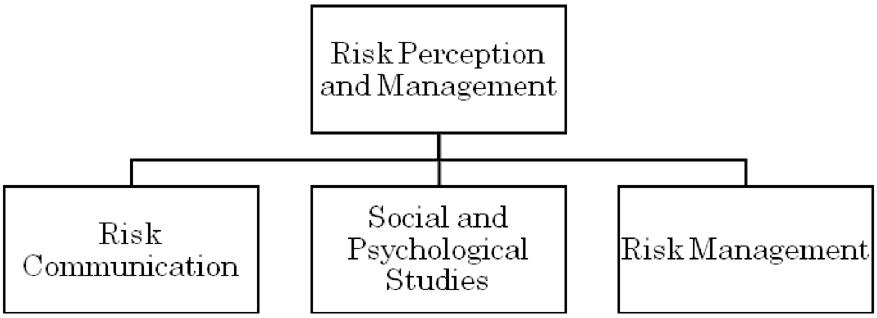


Figure 6. Risk Perception and Management of Microplastics

Another dimension of the proposed management scheme is obtaining feedback from stakeholders on their perception of the risks posed by microplastic contamination (Risk Perception and Management) (Fig. 6). It starts with promoting awareness on the impacts of these pollutants to humans and the environment alike. Educating stakeholders and making them understand the hazards of microplastic contamination is an effective tool for management (Dippo, 2012). Another key aspect of communicating the risks involved is to convert complex terms to a language that can be fully understood by the stakeholders. Developing a management plan for this social, behavioral, and psychological aspect of microplastic management involves surveys, psychological studies, and acknowledging demographic differences. These include regional, cultural, economic, educational, and other variances in behaviour and perceptions (GESAMP, 2015).

A comprehensive management framework is shown in Fig. 7. To mitigate the effects of microplastic contamination to marine and human life, policies should be put into place to strengthen the foundation of the measures to be employed. The illustration also shows the pillars of mitigation action and management which include research, implementation, and regulation. Research entities from the government (Department of Science and Technology, Academic Institutions, National Fisheries Research and Development Institute,) are tasked to develop infrastructure, methods, and technologies that will prevent microplastic entry to the marine environment. Implementation responsibility will fall on the lap of Local Government Units from the Provincial to the Municipal Level headed by the Environmental and Natural Resources Officer to ensure that policies are being implemented using the developed technologies. Regulatory bodies such as the Environmental Management Bureau and Bureau of Fisheries and Aquatic Resources will act as an auditing entity to ensure that effluents and marine bodies are checked in terms of microplastic levels.

## 4. Conclusions

Microplastics were found present in Bacoor bay-cultured tahong (*P. viridis*). This preliminary study confirms a microplastic pollution problem in the Bacoor Bay and Manila Bay area and thus, the need for further research on quantitating and characterizing these pollutants is of utmost importance. Management and mitigation procedures to prevent the entry of microplastics (both primary and secondary) require policy makers, stakeholders, government agencies, non-government organizations, and academic institutions to work in sync to put up a strong and effective management framework with research, implementation, and regulation as its pillars and policy as its foundation.


Figure 6. Risk Perception and Management of Microplastics

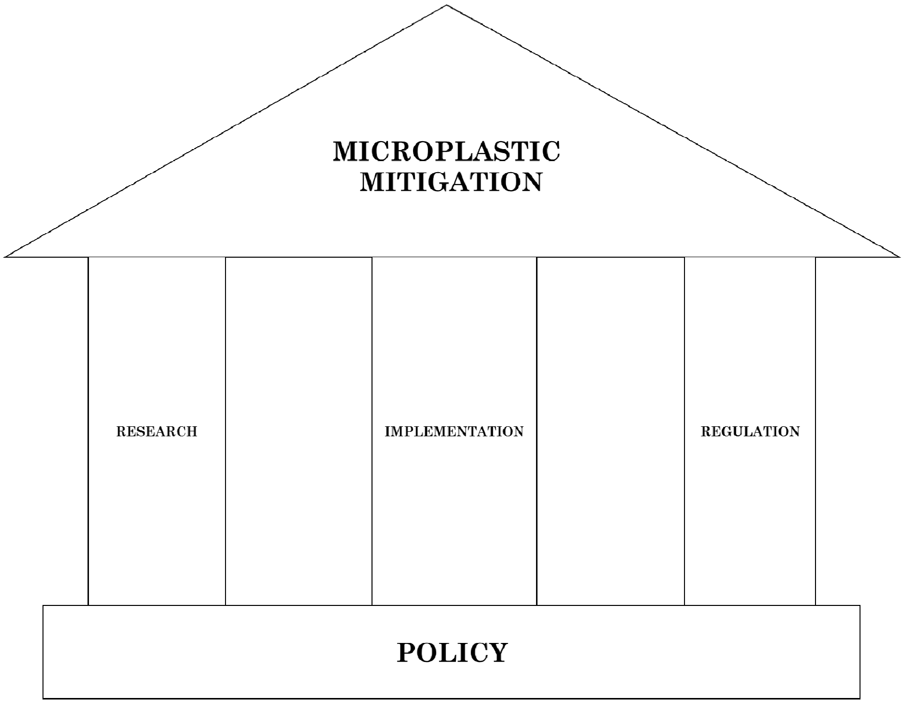


Figure 7. Microplastic Management Framework

## Acknowledgements

The authors would like to thank the Provincial Government of Cavite for assisting in the procurement of the samples and the Biology Laboratory of DLSU for providing the necessary tools and reagents for conducting the analysis.

## References

Andrady AL. Microplastics in the marine environment. Marine Pollution Bulletin 2011; 62(8): 1596-605.

Andrady AL, Neal MA. Applications and societal benefits of plastics. Philosophical Transactions of the Royal Society B 2009; 364(1526): 1977-84.

Chase M, Jones SH, Hennigar P, Sowles J, Harding GCH, Freeman K, Wells PG, Krahforst C, Coombs K, Crawford R, Pederson J, Taylor D. Gulfwatch: monitoring spatial and temporal patterns of trace metal and organic contaminants in the Gulf of Maine (1991-1997) with the Blue Mussel, *Mytilus edulis* L. Marine Pollution Bulletin 2001; 42(6): 491-505.

Dippo B. Microplastics in the coastal environment of West Iceland. Masters Thesis. University Centre of Westfjords, Suorgata 12 400 Isafjorour, Iceland, 2012 [cited 2016 Feb 23]. Available from: http://skemman.is/en.

Figueiras FG, Labarta U, Reiriz MJF. Coastal upwelling, primary production and mussel growth in the Rías Baixas of Galicia. Hydrobiologia 2002; 484: 121-31.

Food and Agricultural Organization (FAO). National Aquaculture Sector Overview (NASO). Viale delle Terme di Caracalla 00153 [homepage on the Internet]. Rome, Italy: 2006; 1: para 28 [cited 2016 Jan 19]. Available from: http://www.fao.org/fishery/countrysector/naso_philippines/en.

GESAMP. Sources, fate and effects of microplastics in the marine environment: a global assessment (*Ed*: Kershaw PJ). Rep. Stud. GESAMP No.90, 96. IMO/FAO/UNESCO IOC/UNIDO/WMO/IAEA/UN/ UNEP/UNDP Joint Group of Experts on the Scientific Aspects of Marine Environmental Protection. 4 Albert Embankment, London SE1 7SR: 2015; 65-68.

Jambeck JR, Geyer R, Wilcox C, Siegler TR, Perryman M, Andrady A, Narayan R, Law KL. Plastic waste inputs from land into the ocean. Science 2015; 347(6223): 768-71.

Masura J, Baker J, Foster G, Arthur C. Laboratory methods for the analysis of microplastics in the marine environment: recommendations for quantifying synthetic particles in waters and sediments. NOAA Technical Memorandum NOS-OR & R-48. NOAA Marine Debris Division 1305 East-West Highway Silver Spring, MD 20910 USA. 2015; 1.

McGuire M, Stevely J. Invasive species of Florida's coastal waters: the Asian Green Mussel (*Perna viridis*) [homepage on the Internet]. Gainesville, FL, USA: 2009; SGEF 175: 1-3 [cited 2016 Feb 12]. Available from: https://edis.ifas.ufl.edu/sg094.

Microbead-Free Waters Act of 2015. Public Law 114-114. 129 Stat. 3129, 2015.

Nor NHMN, Obbard JP. Microplastics in Singapore's coastal mangrove ecosystems. Marine Pollution Bulletin 2014; 79(1-2): 278-83.

Noren F. Small plastic particles in coastal Swedish waters [monograph on the Internet]. Kommunenes Internasjonale Miljøorganisasjon (Local Authorities International Environmental Organisation). Göteborg Sweden: 2007; 6 [cited 2016 Feb 5]. Available from: http://www.kimointernational.org/WebData/Files/Small plastic particles in Swedish West Coast Waters.pdf.

Ocean Conservancy. Stemming the tide: land-based strategies for a plastic-free ocean [monograph on the Internet]. McKinsey Center for Business and Environment. New York, NY 10022: 2015; 7 [cited 2016 Mar 8]. Available from: http://www.oceanconservancy.org/our-work/marine-debris/mckinsey-report-files/full-report-stemming-the.pdf.

Rajagopal S, Venugopalan VP, van der Velde G, Jenner HA. Greening of the coasts: a review of the *Perna viridis* success story. Aquatic Ecology 2006; 40(3): 273-97.

Song YK, Hong SH, Jang M, Han GM, Rani M, Lee J, Shim WJ. A comparison of microscopic and spectroscopic identification methods for analysis of microplastics in environmental samples. Marine Pollution Bulletin 2015; 93(1-2): 202-09.

Thompson RC, Olsen Y, Mitchell RP, Davis A, Rowland SJ, John AWG, McGonigle D, Russell AE. Lost at sea: where is all the plastic?. Science 2004; 304(5672): 838.

Van Cauwenberghe L, Janssen CR. Microplastics in bivalves cultured for human consumption. Environmental Pollution 2014; 193: 65-70.

Van Cauwenberghe L, Devriese L, Galgani F, Robbens J, Janssen CR. Microplastics in sediments: A review of techniques, occurrence and effects. Marine Environmental Research 2015; 111: 5-17.

Vandermeersch G, Van Cauwenberghe L, Janssen CR, Marques A, Granby K, Fait G, Kotterman MJJ, Diogène J, Bekaert K, Robbens J, Devriese L. A critical view on microplastic quantification in aquatic organisms. Environmental Research 2015; 143(Part B): 46-55.

Vasanthi LA, Revathi P, Arulvasu C, Munuswamy N. Biomarkers of metal toxicity and histology of *Perna viridis* from Ennore estuary, Chennai, southeast coast of India. Ecotoxicology and Environmental Safety 2012; 84: 92-98.

Vegter AC, Barletta M, Beck C, Borrero J, Burton H, Campbell ML, Costa MF, Eriksen M, Eriksson C, Estrades A, Gilardi KVK, Hardesty BD, Ivar do Sul JA, Lavers JL, Lazar B, Lebreton L, Nichols WJ, Ribic CA, Ryan PG, Schuyler QA, Smith SDA, Takada H, Townsend KA, Wabnitz CCC, Wilcox C, Young LC, Hamann M. Global research priorities to mitigate plastic pollution impacts on marine wildlife. Endangered Species Research 2014; 25: 225-47.

Yap W. Rural Agriculture in the Philippines. Food and Agriculture Organization for the United Nations (Regional Office for Asia and the Pacific). Bangkok, Thailand. 1999; 2: 7.

*Received 31 March 2016*
*Accepted 28 April 2016*

**Correspondence to**
Associate Professor Dr. Jose Isagani B. Janairo
Biology Department, College of Science,
De La Salle University,
2401 Taft Avenue,
Manila 0922
Philippines
E-mail: jose.isagani.janairo@dlsu.edu.ph