

Volume 220

David M. Whitacre *Editor*

Reviews of Environmental Contamination and Toxicology

With Cumulative and Comprehensive
Index of Subjects Covered

Volumes 211-220

 Springer

Reviews of
Environmental Contamination
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VOLUME 220

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ISSN 0179-5953

ISBN 978-1-4614-3413-9

e-ISBN 978-1-4614-3414-6

DOI 10.1007/978-1-4614-3414-6

Springer New York Dordrecht Heidelberg London

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Printed on acid-free paper

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Foreword

International concern in scientific, industrial, and governmental communities over traces of xenobiotics in foods and in both abiotic and biotic environments has justified the present triumvirate of specialized publications in this field: comprehensive reviews, rapidly published research papers and progress reports, and archival documentations. These three international publications are integrated and scheduled to provide the coherency essential for nonduplicative and current progress in a field as dynamic and complex as environmental contamination and toxicology. This series is reserved exclusively for the diversified literature on “toxic” chemicals in our food, our feeds, our homes, recreational and working surroundings, our domestic animals, our wildlife, and ourselves. Tremendous efforts worldwide have been mobilized to evaluate the nature, presence, magnitude, fate, and toxicology of the chemicals loosed upon the Earth. Among the sequelae of this broad new emphasis is an undeniable need for an articulated set of authoritative publications, where one can find the latest important world literature produced by these emerging areas of science together with documentation of pertinent ancillary legislation.

Research directors and legislative or administrative advisers do not have the time to scan the escalating number of technical publications that may contain articles important to current responsibility. Rather, these individuals need the background provided by detailed reviews and the assurance that the latest information is made available to them, all with minimal literature searching. Similarly, the scientist assigned or attracted to a new problem is required to glean all literature pertinent to the task, to publish new developments or important new experimental details quickly, to inform others of findings that might alter their own efforts, and eventually to publish all his/her supporting data and conclusions for archival purposes.

In the fields of environmental contamination and toxicology, the sum of these concerns and responsibilities is decisively addressed by the uniform, encompassing, and timely publication format of the Springer triumvirate:

Reviews of Environmental Contamination and Toxicology [Vol. 1 through 97 (1962–1986) as Residue Reviews] for detailed review articles concerned with any aspects of chemical contaminants, including pesticides, in the total environment with toxicological considerations and consequences.

Bulletin of Environmental Contamination and Toxicology (Vol. 1 in 1966) for rapid publication of short reports of significant advances and discoveries in the fields of air, soil, water, and food contamination and pollution as well as methodology and other disciplines concerned with the introduction, presence, and effects of toxicants in the total environment.

Archives of Environmental Contamination and Toxicology (Vol. 1 in 1973) for important complete articles emphasizing and describing original experimental or theoretical research work pertaining to the scientific aspects of chemical contaminants in the environment.

Manuscripts for Reviews and the Archives are in identical formats and are peer reviewed by scientists in the field for adequacy and value; manuscripts for the *Bulletin* are also reviewed, but are published by photo-offset from camera-ready copy to provide the latest results with minimum delay. The individual editors of these three publications comprise the joint Coordinating Board of Editors with referral within the board of manuscripts submitted to one publication but deemed by major emphasis or length more suitable for one of the others.

Coordinating Board of Editors

Preface

The role of *Reviews* is to publish detailed scientific review articles on all aspects of environmental contamination and associated toxicological consequences. Such articles facilitate the often complex task of accessing and interpreting cogent scientific data within the confines of one or more closely related research fields.

In the nearly 50 years since *Reviews of Environmental Contamination and Toxicology* (formerly *Residue Reviews*) was first published, the number, scope, and complexity of environmental pollution incidents have grown unabated. During this entire period, the emphasis has been on publishing articles that address the presence and toxicity of environmental contaminants. New research is published each year on a myriad of environmental pollution issues facing people worldwide. This fact, and the routine discovery and reporting of new environmental contamination cases, creates an increasingly important function for *Reviews*.

The staggering volume of scientific literature demands remedy by which data can be synthesized and made available to readers in an abridged form. *Reviews* addresses this need and provides detailed reviews worldwide to key scientists and science or policy administrators, whether employed by government, universities, or the private sector.

There is a panoply of environmental issues and concerns on which many scientists have focused their research in past years. The scope of this list is quite broad, encompassing environmental events globally that affect marine and terrestrial ecosystems; biotic and abiotic environments; impacts on plants, humans, and wildlife; and pollutants, both chemical and radioactive; as well as the ravages of environmental disease in virtually all environmental media (soil, water, air). New or enhanced safety and environmental concerns have emerged in the last decade to be added to incidents covered by the media, studied by scientists, and addressed by governmental and private institutions. Among these are events so striking that they are creating a paradigm shift. Two in particular are at the center of ever-increasing media as well as scientific attention: bioterrorism and global warming. Unfortunately, these very worrisome issues are now superimposed on the already extensive list of ongoing environmental challenges.

The ultimate role of publishing scientific research is to enhance understanding of the environment in ways that allow the public to be better informed. The term “informed public” as used by Thomas Jefferson in the age of enlightenment conveyed the thought of soundness and good judgment. In the modern sense, being “well informed” has the narrower meaning of having access to sufficient information. Because the public still gets most of its information on science and technology from TV news and reports, the role for scientists as interpreters and brokers of scientific information to the public will grow rather than diminish. Environmentalism is the newest global political force, resulting in the emergence of multinational consortia to control pollution and the evolution of the environmental ethic. Will the new politics of the twenty-first century involve a consortium of technologists and environmentalists, or a progressive confrontation? These matters are of genuine concern to governmental agencies and legislative bodies around the world.

For those who make the decisions about how our planet is managed, there is an ongoing need for continual surveillance and intelligent controls to avoid endangering the environment, public health, and wildlife. Ensuring safety-in-use of the many chemicals involved in our highly industrialized culture is a dynamic challenge, for the old, established materials are continually being displaced by newly developed molecules more acceptable to federal and state regulatory agencies, public health officials, and environmentalists.

Reviews publishes synoptic articles designed to treat the presence, fate, and, if possible, the safety of xenobiotics in any segment of the environment. These reviews can be either general or specific, but properly lie in the domains of analytical chemistry and its methodology, biochemistry, human and animal medicine, legislation, pharmacology, physiology, toxicology, and regulation. Certain affairs in food technology concerned specifically with pesticide and other food-additive problems may also be appropriate.

Because manuscripts are published in the order in which they are received in final form, it may seem that some important aspects have been neglected at times. However, these apparent omissions are recognized, and pertinent manuscripts are likely in preparation or planned. The field is so very large and the interests in it are so varied that the editor and the editorial board earnestly solicit authors and suggestions of underrepresented topics to make this international book series yet more useful and worthwhile.

Justification for the preparation of any review for this book series is that it deals with some aspect of the many real problems arising from the presence of foreign chemicals in our surroundings. Thus, manuscripts may encompass case studies from any country. Food additives, including pesticides, or their metabolites that may persist into human food and animal feeds are within this scope. Additionally, chemical contamination in any manner of air, water, soil, or plant or animal life is within these objectives and their purview.

Manuscripts are often contributed by invitation. However, nominations for new topics or topics in areas that are rapidly advancing are welcome. Preliminary communication with the editor is recommended before volunteered review manuscripts are submitted.

Summerfield, NC, USA

David M. Whitacre

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Plastics in the Marine Environment: The Dark Side of a Modern Gift

Jort Hammer, Michiel H.S. Kraak, and John R. Parsons

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1 Introduction

Plastics are one of the most widely used materials in the world; they are broadly integrated into today's lifestyle and make a major contribution to almost all product areas. The typical characteristics that render them so useful relate primarily to the fact that they are both flexible and durable. These characteristics are very useful when plastics are used in everyday life. But when plastics are discarded into the environment they can persist for very long periods of time. Because of their nearly indestructible morphology and the toxins they contain, plastics can seriously affect ecosystems (UNEP 2005).

The biggest mass of plastic debris occurs in the oceans' major gyres (Moore et al. 2001). Herein, the rotation of ocean currents catches any sea debris that floats and moves it to the vortex center, where it accumulates. Currently, the plastic debris patch in the North Pacific Ocean covers an area as large as France and Spain together. This debris constitutes particles that have diameters as small as several millimeters to big plastic-filled "ghost nets" having a weight of 2,000 kg. This debris affects all ocean life, and because we are at the top of the food chain, it affects humans too.

The aim of this review is to address and answer the following questions from information sourced largely from scientific reports and the mainstream scientific literature: What are plastics actually? What happens when they are discarded? How do plastics pose a threat to organisms in marine environments, and what are the solutions to the plastic debris problem?

2 Facts About Plastics

2.1 History of Plastic

The term plastics comes from the Greek word "plastikos" meaning "fit for molding," and refers to the plasticity of these materials during their manufacture (Liddell et al. 1968). Nowadays, plastics¹ is the term applied to a wide range of synthetic organic compounds that are produced by polymerization, and these consist of many repeating units (monomers) that come together to create copolymers. The plasticity of plastics allows them to be pressed or extruded into many different shapes and forms. Because of their sometimes infinitely long molecular structures, they can be very flexible and strong.

Plastics have been developed to replace depleted natural resources since ancient times. Polymers were used in 1600 B.C. by the ancient Mesoamericans, the first to

¹The term *plastics* refers to a large number of synthetic organic compounds that have a polymeric structure and the ability to be cast in various shapes. However, the term *plastic* only refers to the plasticity of a material.

process natural rubber, to make figurines and bands (Hosler et al. 1999). Several semisynthetic plastics like polystyrene (PS) and polyvinyl chloride (PVC) were discovered in the nineteenth century, which marks the beginning of the plastic era (Ebewele 2000). Initially, plastics could not be used in commercial products because of their often rigid and brittle structure. This changed in 1909, when the first true synthetic phenol-formaldehyde plastic material (Bakelite) was discovered and was used in many different products, from telephone handsets to engine parts (Groot 2009). Later, in 1926, the modern form, PVC, was created as a plasticized polyvinyl chloride (vinyl), and in 1933 polyvinylidene chloride (PVDC), or Saran, was introduced by Ralph Wiley (Morris 1986).

Polyurethane (PUR), a flexible foam, was invented in 1937. In 1938, polystyrene (PS) became commercially practical and was used in peanut packaging; in this same year, polytetrafluoroethylene (PTFE) or Teflon was invented by Roy Plunkett. In 1939, nylon and Neoprene were invented by Wallace Carothers. Polyethylene terephthalate (PET), also known as polyester, was introduced by John Rex Whinfield in 1941. Polyester is primarily used in the manufacture of beverage bottles (PackagingToday 2009).

World War II increased the worldwide demand for plastics because copper, aluminum, and steel became so valuable for military use. Thereafter, plastics quickly gained use as a manufacturing material, and consequently material manufacturers, machine builders, and mold-makers flourished (Beall 2009). After the Second World War ended, civilian outlets were needed for plastics to keep the factories in business. The market was rapidly overwhelmed with plastic products, which were regarded by society to be “cheap and disposable.” In 1979, the plastic production in the USA exceeded that of steel production. Hence, one could conclude that World War II changed the world and started the age of the plastic industry (Beall 2009; Morris 1986).

In 1951, high-density polyethylene (HDPE) and polypropylene (PP) were invented and were employed for use in making water jugs and hula hoops. In 1954, Styrofoam was invented. Styrofoam is a trademark for extruded polystyrene foam and weighs 30-fold less than normal polystyrene foam. Thermoplastic polyester, which is based on polybutylene terephthalate (PBT), was introduced in 1970. This thermoplastic polymer is used as a material for high-quality, highly stressed engineering parts in many industrial sectors as a result of its high strength and good stability at high temperatures (Beall 2009).

2.2 *Production*

Plastics are produced by the conversion of natural products or by synthesis from primary chemicals, generally from oil, natural gas, or coal (Morris 1986; Thompson et al. 2009b). After conversion by a compounder fabric, the plastics become building materials for thousands of plastic products that are used worldwide. The fabrics, which give shape to plastics and are used to produce plastic products, are called “converters.” The most economical way to ship large quantities of a solid material

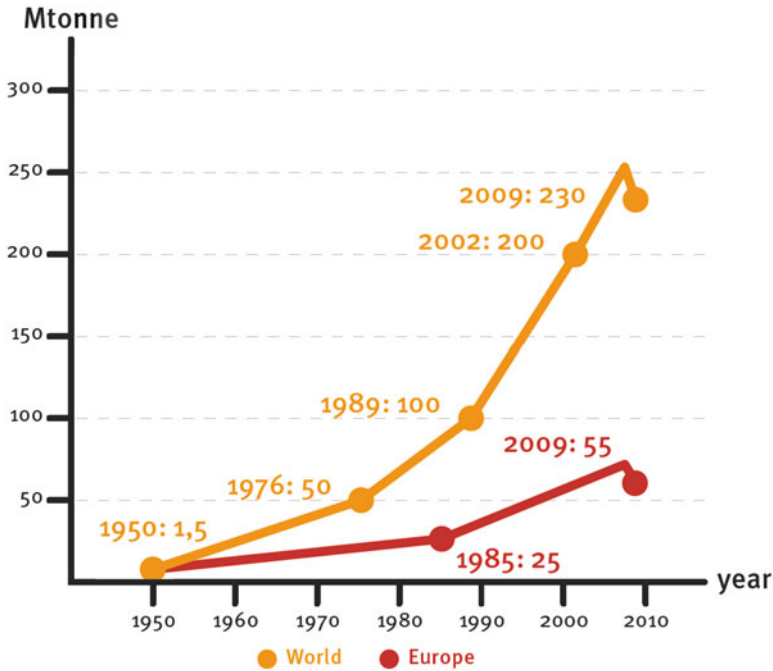


Fig. 1 World plastics production from 1950 to 2009 in millions of tons (PEMRG 2010)

from a compounder to these converters is in pelletized form (Ogata et al. 2009). Plastic-producing manufacturers utilize a form of preproduction pellets that are called “nurdles.” Nurdles are about 5 mm in diameter and weight approximately 20 mg each. After production, the nurdles are shipped to converters by rail tank cars which contain around one billion nurdles per tanker. In the USA, approximately 27 million tons of nurdles are produced annually, which constitute 1.35 quadrillion granules (EPA 1993). These preproduction nurdles can be subjected to different manufacturing processes to produce different products (Andrady 2003).

Once plastics became components of building materials that were commercially used in products and in the construction industry, their production and consumption increased significantly. The global production of plastics between 1950 and 2009 showed an average annual increase of 9%. In 1950, 1.5 million tons of plastics were produced and this has increased to 230 million tons in 2009 (Fig. 1). In 2008, the production dropped by 15 million tons as a consequence of the global financial crisis (Gioia et al. 2008). In mid-2009, there were signs of a market recovery, and in 2010 the annual production increased to 265 million tons (PEMRG 2011). The current plastic consumption per capita has grown to approximately 100 kg/year for NAFTA (North American Free Trade Agreement, including Canada, the USA, and Mexico) and Western Europe. If the growth continues, projected consumption will become 140 kg per capita in 2015. The biggest potential growth is expected from rapidly developing countries in Asia and the new European member states.

2.3 Additives

Plastics can be modified by adding a variety of chemicals (additives) that impart specific properties for the end product. Additives are specific chemical compounds that are added to a basic polymer to alter or improve its properties. The use of common plastics in today's products would not be possible without the use of such additives. PVC, for example, is very sensitive to thermal- and photo-degradation and is not useful without the addition of stabilizer additives, such as antioxidants and UV stabilizers (ACC 2010; Andrady 2003). Some of these additives, however, may cause a variety of toxic effects. For example, flame retardants (e.g., polybrominated diphenyl ethers), which are often added to plastics like PVC, can leach from food packaging's into food and are suspected to be endocrine disruptors (Hale et al. 2002). Phthalates are a widely used group of plasticizing chemicals that are primarily utilized in PVC polymers. Di-2-ethylhexyl phthalate (DEHP) is the major plasticizer used in medical devices such as blood bags, catheters, and tubing (Koch and Calafat 2009). The primary building block of polycarbonate, bisphenol A, is known to be an endocrine disruptor, and is often used in food packaging (Nadal et al. 2009). Toxic metals such as lead (McIlgorm et al. 2011) and chromium (Cr) can also be present in polymers. These metals are often used in pigments that are added to plastics, and are potentially released into the environment (Omori et al. 2011). The toxicity of plastics and their additives is further discussed in Sect. 4.3.

3 Plastic Debris in the Marine Environment

3.1 Introduction

Plastics are often light, cheap, and durable materials. Because they can usually be cheaply produced, they are generally used only once and are then thrown away as litter. The fact that plastics are light and durable causes such litter to accumulate in landfills, or to be transported from source areas to sinks like the ocean. About 49% of all produced plastics are buoyant, which gives them the ability to float, and thereby travel on ocean currents to anyplace in the world (EPA 2008). As addressed below, a good understanding of the transport and fate of plastics in the ocean can be gained by categorizing and monitoring the movement of plastic debris.

3.2 Categorization of Plastic Debris

Plastic debris in the environment is routinely monitored to gain insights that concern the quantity and geography of its distribution. To this purpose, plastic debris is divided into three classes: macrodebris (>20 mm diameter), mesodebris (2-20 mm), and microdebris (<2 mm) (Galgani and Lecornu 2004; Thompson et al. 2009b), although some authors recommend other size limits (Cheshire et al. 2009).

3.2.1 Macrodebris

Macrodebris relates to the larger parts of plastic debris (>20 mm to several meters). Large-sized plastic debris may comprise plastic chairs, shoes, car/plane/boat parts, buoys, footballs, etc. Nearly any object larger than 20 mm that has ever been made from plastic is found in the oceans. An important, often found piece of macrodebris is the “ghost net.” A ghost net is an abandoned or lost fishing net that roams the ocean. A ghost net travels with the currents and tides, continually catching animals and other macrodebris in its maze, and becomes filled primarily with other plastic objects. Ghost nets can grow to masses of 6 ton, and are often too heavy and too large to be removed from the ocean (CGNP 2009).

3.2.2 Mesodebris

Mesodebris often consists of plastic resin pellets, also known as nurdles. Nurdles are small granules that have the shape of a cylinder or disk, and have a maximum diameter of 5 mm. The pellets are made as raw industrial material, and are sent to manufacturers for remelting and molding into plastic products (Ogata et al. 2009). Because of their small size, nurdles are often accidentally expelled into the environment during transport and manufacturing. They then travel by surface run-off, rivers, and streams toward the ocean. Nurdles are highly persistent, and therefore are widely distributed in the ocean, and are found on beaches and water surfaces all over the world (Barnes et al. 2009; Derraik 2002; Edyvane et al. 2004; Ogata et al. 2009).

3.2.3 Microdebris

Microdebris consists of small plastic fragments <5 mm in diameter. Meso- and macro-debris can fragmentize into smaller bits from the constant movement and collisions with other plastic debris, or from the influence of UV-radiation and photo-oxidative degradation (Ng and Obbard 2006; Shaw and Day 1994). These microdebris fragments can become as small as 2 μm . Other small plastic particles, also called “scrubbers,” which originate from hand cleaners, cosmetic products, and airblast cleaning media, have also contaminated the marine environment. Scrubbers are often contaminated with other chemicals (see Sect. 4.3) and can easily be ingested by filter-feeding organisms (Fendall and Sewell 2009; Gregory 1996).

3.3 *Origin of Plastics in the Marine Environment*

The release of plastics into the environment is a result of inappropriate waste management, improper human behavior, or incidental pollution (Barnes et al. 2009). Well-operated landfills are closed systems; they are daily covered by soil or synthetic

materials and are surrounded by fences to hold wind-blown debris in place. Plastics do not biodegrade and can remain in place for centuries, until they are burned or used for recycling. The portion of plastic litter that does not reach landfills will roam the earth's surface, travelling by wind until it reaches the rivers, and eventually the sea. Improper human behavior produces such waste, when plastics are abandoned or are dumped outside licensed collection points or at sea. Incidental pollution also occurs, and includes the loss of containers at sea (Barnes et al. 2009).

In highly populated areas, land-based sources dominate the input of plastic waste into the marine environment; ship-generated debris is the major source of marine debris found on remote shores. The US Academy of Sciences estimated the total annual input of marine debris into the oceans to be approximately 6.4 million tons. Furthermore, eight million items of marine litter are estimated to enter the oceans and seas every day through various sources (UNEP 2005; 2009b).

3.3.1 Ocean-Based Sources

Nearly 5.6 million tons of marine debris every year is estimated to come from ocean-based sources, which is 88% of the total marine debris input. Daily, about five million items of solid marine debris are estimated to be thrown overboard or lost from ships (UNEP 2009b). The main ocean-based sources of such waste are as follows (Sheavly 2005; UNEP 2001; 2009b).

Merchant Ships, Ferries, and Cruiseliners

These ships are sources for marine debris in the form of household waste, sewage, cargo, and cargo hold waste (wiring straps, covering material and cargo residues), packaging material (plastic sheets, boxes), engine-room waste (oil or detergent containers), and discarded medical and sanitary equipment. The debris is intentionally dumped for lack of sufficient storage facilities or because of negligence, and sometimes is lost accidentally through careless handling or bad weather.

Naval and Research Vessels

Naval and research vessels produce much of the same garbage as do the merchant ships, ferries, and cruiseliners, but military vessels may also deliberately dump military items to dispose of them. An example of this is the dumping of old military equipment in the Marsdiep by the Dutch Navy.

Pleasure Craft

From these craft, primarily household waste, sewage waste, oil containers, and recreational fishing gear (angling line and weights) are dumped from ignorance, negligence, or lack of reception facilities in local harbors.

Table 1 A summary of abandoned/discarded and lost polymer-containing fishing gear from around the world (taken from articles summarized by UNEP 2009b)

Region	Fishery/gear type	Indicator of gear loss
North Sea and NE Atlantic	Bottom-set gillnets	0.02–0.09% nets lost per boat per year
English Channel and North Sea (France)	Gillnets	0.2% (sole and plaice) to 2.11% (sea bass) nets lost per boat per year
Mediterranean	Gillnets	0.05% (inshore hake) to 3.2% (sea bream) nets lost per boat per year
Gulf of Aden	Traps	20% lost per boat per year
United Arab Emirates Sea Area	Traps	260,000 lost per year in 2002
Indian Ocean	Maldives tuna longline	3% loss of hooks/set
Australia (Queensland)	Blue swimmer crab trap fishery	35 traps lost per boat per year
NE Pacific	Bristol Bay king crab trap fishery	7,000–31,000 traps lost in the fishery per year
NW Atlantic	Newfoundland cod gillnet Fishery	5,000 nets per year
	Canadian Atlantic gillnet Fisheries	2% nets lost per boat per year
	Gulf of St Lawrence snow crab	792 traps per year
	Net England lobster fishery	20–30% traps lost per boat per year
	Chesapeake Bay	30% traps lost per boat per year
Caribbean	Guadeoupe trap fishery	20,000 traps lost per year

Offshore Oil or Gas Platforms

Drill pipes and drill pipe protectors, hard hats, cotton and rubber gloves, storage drums, oil containers household waste, discarded medical and sanitary equipment are lost from offshore platforms. The waste is usually dumped on purpose and sometimes is accidentally lost from careless handling or bad weather.

Fishing Vessels

Most ocean-based marine litter is probably represented by abandoned and lost fishing gear. In areas far away from urban development, discarded fishing gear is responsible for 50–90% of the total marine debris. Table 1 shows a summary of the types of abandoned, discarded, or lost gear that reaches the oceans around the world every year. Among the different forms of discarded marine debris from fishing vessels are fishing nets, fishing lines, fish boxes, crab and lobster pots, oyster nets, strings for packaged bait, rubber gloves and of course household waste, oil containers, and sewage. There are several reasons as to why fishing gear can become marine litter (UNEP 2009b):

– *Fishing gear is abandoned*

Some fishing gear and nets are abandoned by their owners and are never retrieved after falling into the ocean. This generally happens when fishing activities are

illegal, unregistered, and unreported; illegal gear is often abandoned because fishing vessels cannot enter a harbor and be seen with this equipment, or to avoid inspections when fishing occurs in forbidden areas. Finally, abandonment may result from the lack of time to collect all nets or traps.

– *Fishing gear is discarded*

Fishing gear is often discarded when damaged; it is often cheaper to discard a damaged item, than to transfer the gear for onshore disposal. This occurs for many discarded and dumped marine debris items; it is cheaper and faster to dump everything overboard than to arrange for onshore disposal.

– *Fishing gear is lost*

Accidental loss of fishing gear at sea often happens due to gear conflict (nets from different vessels become entangled with each other), misplaced gear, poor topography (nets and traps become struck on the seafloor), and extreme weather.

– *Containers are lost*

Between 1990 and 2005, 16,625 containers worldwide were reported as lost by the Institute of Shipping Economics and Logistics (ISL 2009). Transport containers can contain several thousand pairs of shoes, televisions, or rubber ducks (Ebbesmeyer and Ingraham 1994); these are generally buoyant, and therefore the container may open and discharge contents when waterlogged. The loss of containers at sea is primarily caused by heavy weather (42%) and collisions between ships (11%). Since the fleet of container ships has grown by 140% since 1994, the chance of losing containers has increased accordingly (ISL 2009).

3.3.2 Land-Based Sources

Approximately 0.8 million tons annually of marine debris, which is 12% of the total debris input into the oceans, originates from land-based sources, and primarily consists of discarded plastic items (user plastic). In highly populated areas, marine debris comes primarily from the land. Main land-based sources of marine debris are as follows (Sheavly 2005; UNEP 2005; 2009b).

Municipal Landfills Located on the Coast

Many poorly managed or illegal landfills on the coast contribute to marine debris (solid household waste) under the influence of wind, which blows litter into the sea, or from flooding of the landfill area.

Transport of Waste by Rivers from Landfills, or Any Other Sources of Debris Along River- and Waterway Systems

Solid household waste and other items are flushed into the river after water levels rise, or from the influence of heavy rains. Debris could also be blown into rivers or

illegally dumped (Moore et al. 2005). Moore et al. (2005) quantified the contribution of plastic particles from two rivers draining a large urban area (Los Angeles). Samples were taken from different depths in the rivers, and from one moderate and one heavy rain day, and one dry day. A total of 72 h of monitoring by using a net resulted in collecting a total of 2,333,871,120 (2.3 billion) plastic objects and fragments having a total weight of 30,438.52 kg (Moore et al. 2011).

Discharge of Untreated Sewage and Storm Water

In many of the world's cities, untreated sewage and storm water is discharged into the rivers and into the sea. Storm water carries solid and liquid items that are thrown onto streets and are subject to being washed away.

Industrial Facilities

The enormous amount of plastic resin pellets found in the sea today originates from industrial facilities. Also, untreated waste water from landfills delivers a large mass of solid material into the sea. Other materials, which originate from industrial facilities, are packaging material and production scrap.

Tourism

Various kinds of food packages, beverage cans and cartons, toys, and cigarettes are left at the beach by numerous tourists. This debris often blows into the sea or is taken off shore by the tide.

In summary, most plastic debris originates from ocean-based sources such as waste from cruise ships or fishing gear from the fishing industry. Land-based plastic debris is often only found near highly populated areas.

3.4 Degradation of Plastics in the Marine Environment

Most polymeric materials that enter the environment are subjected to degradation² that is caused by a combination of factors, including thermal oxidation, photo-oxidative degradation, biodegradation, and hydrolysis. The common plastics found

²Degradation implies here to the loss of useful properties following chemical changes in polymeric materials. When plastic material is technically said to be fully degraded, the polymer structure no longer exists (Andrady 1994).

in marine environments, however, do not biodegrade and primarily break down through photo-oxidative degradation. Furthermore, unlike plastics exposed on land, exposed plastics floating on the ocean's surface do not suffer from heat build-up due to absorption of infrared radiation, and therefore barely undergo thermal oxidation (Andrady 1994, 2003; Andrady et al. 1993). The degradation of negatively buoyant plastics depends on very slow thermal oxidation, or hydrolyses, as a result of most wavelengths being readily absorbed by water. Hence, plastics residing in marine environments degrade at a significantly slower rate than they do on land. Biodegradable plastics will further be discussed in Sect. 5.

Plastics primarily break down through photo-oxidative degradation, which is activated by solar radiation. The spectral energy of solar radiation reaching the earth's surface ranges between 298 nm in the ultraviolet (UV) region and 2,500 nm in the near-infrared region. Because short wavelengths contain more photonic energy than long wavelengths, short wavelengths have a stronger actinic³ effect on materials and are capable of breaking strong bonds. Therefore, most photo-oxidative degradation occurs in the UV wavelength range of solar radiation (298–420 nm). However, regardless of the intensity, a specific wavelength can only cause damage to a surface when the material is capable of absorbing the specific wavelength. Thus, the effect of solar radiation on plastic depends on (1) the wavelength and amount of radiation a polymer is able to absorb and (2) the strength of the chemical bonds within the polymer (Andrady 2003).

The direct absorbance of solar radiation by a polymer is often determined by the presence of chromophores⁴, which can absorb wavelengths longer than 290 nm. Only aromatic polymers like polyarylate (PAR) and PET contain structural chromophores capable of absorbing UV radiation. Nonaromatic (aliphatic) polymers, like polyethylene and PVC, do not contain chromophores and their UV absorbance lies below the range of the spectral energy of solar radiation. However, most aliphatic plastics contain solar UV absorbing impurities like catalyst residues, organic contaminants, and thermal oxidation products attached to the polymer chain, which makes them sensitive to photo-oxidation. A small amount of radiation absorbed by these impurities can initiate a rapid free-radical⁵ chain reaction that can cause extensive photo-oxidation. This reaction causes many aliphatic polymers to be indirectly more sensitive to radiation than aromatic polymers, while the latter are able to directly absorb much more solar UV radiation (Andrady 2003; Hamidi 2000).

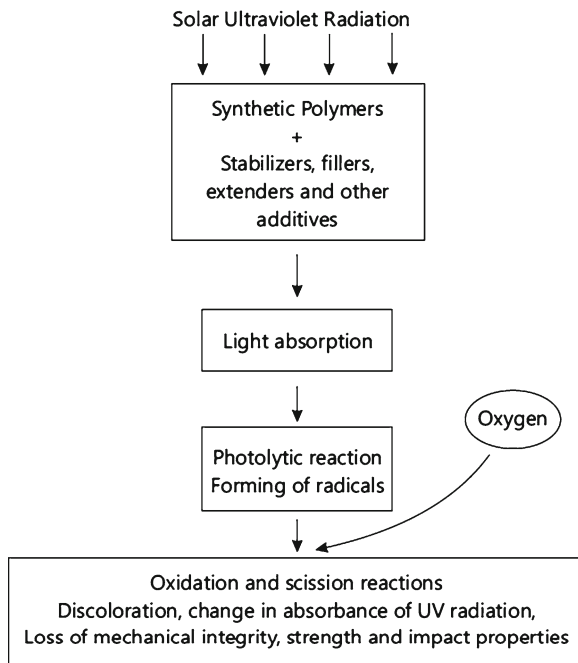
Two major reaction mechanisms occur by which solar radiation can degrade polymer materials: (1) a reaction is initiated by photolysis of the chromophores as a result of absorbing UV radiation, which produces a hydroxy radical, and (2) a photo-oxidative chain reaction is initiated by the energy absorbed by impurities. The radicals

³ Actinism is the intrinsic property in radiation that produces photochemical activity.

⁴ A chromophore is a chemical group capable of selective light absorption resulting in the coloration of certain aromatic organic compounds.

⁵ A free-radical is a usually short-lived atom or molecule with at least one unpaired electron. Free-radicals are often highly reactive and unstable.

Fig. 2 A schematic diagram of polymer degradation under the influence of UV radiation (adapted from Andradý et al. 1998)



created by these two pathways react with oxygen and the polymer to produce cross-link bonds. Therefore, the polymer loses tensile strength, elasticity, and stretch; it becomes more brittle and breaks more easily (Andradý 2003; Andradý et al. 1998). Photolysis of the chromophores reduces coloration and thus causes bleaching of the polymeric material (Shaw and Day 1994) (Fig. 2). Synthetic polymers are only degraded by solar UV radiation of which the UV spectrum constitutes only 1%; therefore, degradation of polymers is a long-lasting process. Annually, ever larger amounts of plastic debris are introduced into the environment than can be degraded. Therefore, plastics are constantly accumulating in the oceans and in coastal areas.

It was shown in a recent study (Sivan 2011) that biodegradation of plastic waste could be possible with selected microbial strains. By incorporating pro-oxidants (photo sensitizers) into the polymer chain, a photochemical reaction can quickly be initiated via the catalytic activity of these oxides. This reaction causes oxidative degradation of the polymeric molar mass and forms oxygenated groups (such as carbonyl), which are then more easily metabolized by microorganisms. Although degradation of plastics would still be a long lasting process, microbes would speed up the process; e.g., after 1 year of natural weathering and 3 months of composting at 58°C. Twelve percent of the original carbon present in test samples were microbially mineralized (Sivan 2011).

3.5 Accumulation of Plastics in the Marine Environment

The persistence of plastics causes them to accumulate in the environment. The mass production of plastics started in the 1950s. Today, marine debris is dominated by plastics. It is estimated that half a century ago the amount of anthropogenic debris in the ocean would have been four orders of magnitude lower than it is today (UNEP 2005). The percentage of plastic fragments that exists in marine debris increases as the distance from the debris source increases. This characteristic is caused by the low weight and strength of plastics, which renders them easily transported further than other debris, resulting in plastic contamination, even in the most remote places on earth. Plastic objects are primarily found floating on the sea surface or along shorelines where they have been washed ashore. Research in the North Sea showed that, of all plastic debris annually dumped in the sea, 15% is floating on the surface, 15% is washed ashore, and eventually, 70% will sink to the sea bottom (Barnes et al. 2009; UNEP 2001).

3.5.1 Floating Plastic Debris

Many plastic items float, because they consist of light polymeric material, or because their shapes allow them to trap air (e.g., bottles and bags). Most plastic objects float until they either become too heavy from biota growing on their surface, or because they become waterlogged and sink.

Monitoring Floating Debris

The abundance of floating plastic at sea can be estimated by observing large plastic items or by using net trawls to collect smaller items. The success of visual observations depends on the number of observers. Rather large areas can be scanned for debris, especially when aerial observation is performed. Less subjective observations are made by using net trawls, but these are limited to sampling smaller areas. Most net trawl samples are taken with a manta trawl, a device which captures surface debris in a fine mesh net. A manta trawl has a 90-cm wide opening, with a small collection sock attached to it, which consists of a 0.333-mm mesh net. Another way to sample is with a 3-m long and 1-cm wide bongo net. This net also consists of a 0.333-mm mesh size and can be used to take samples from 10 to 100 m depths (AMRF 2010; Ryan et al. 2009).

Plastics Accumulation at Sea

Floating debris appears to particularly accumulate in oceanographic convergence areas, enclosed seas, and ocean currents. The North Pacific central gyre, an area of high atmospheric pressure with a clockwise ocean current, forces debris into a

central area where winds and currents fade away. This gyre has been widely used for sampling and investigating plastic debris. Meanwhile, because of the inexorable accumulation of plastic debris, mostly meso- and micro-plastic particles, the center of the North Pacific gyre is now known as the Great Pacific “Garbage Patch” or “Pacific Trash Vortex” (Allsopp et al. 2007).

Moore et al. (2001) used a manta trawl to sample 11 random sites in the eastern area of the North Pacific central gyre. The individual plastic pieces collected were segregated by type into five categories: unidentified fragments, Styrofoam fragments, plastic resin pellets, polypropylene (sailboat) line fragments, and thin plastic film fragments. The mean abundance of plastic particles in the surveyed area was 334,271 particles/km² with a mass of 5,114 g/km². The abundance of plankton was measured to be five times higher than that of plastic, but the mass of the plastic particles was approximately six times that of plankton (Moore et al. 2001). In 2002, paired bongo nets were used to sample another area in the eastern part of the North Pacific central gyre. The nets were brought to a depth of 10 and 30 m. The samples collected at both depths contained a mean particle density of 0.017 particles/m³, a factor 100 lower than densities found at the surface of the same sites that were sampled earlier (Moore et al. 2005).

Another undertaking to observe plastic particle density in the ocean was performed at the western side of the North Pacific gyre in the Kuroshio Current area (Yamashita and Tanimura 2007). Here, between April 2000 and April 2001, 76 locations were sampled using a manta trawl. Plastics were categorized as follows: plastic resin pellets, plastic products, fragments of plastic products, rubber, fiber, Styrofoam, plastic sheets (less than 2 mm thick), and sponge. The abundance (0–3,520,000 particles/km²) and mass (0–153,000 g/km²) varied among the locations. The abundance of plastic particles increased as distance from the shore increased, and the maximum abundance occurred in the area of the Kuroshio Current, which implies that this current plays a role in the transport and distribution of plastics from Japan and Indonesia over the North Pacific Ocean (Yamashita and Tanimura 2007).

The North Pacific gyre is only one of five gyres that are present on earth. The North Atlantic gyre has also been investigated and research institutions have been working on mapping their data. The Sea Education Association (SEA) monitored the North Atlantic gyre for plastics between 1986 and December 2008. More than 6,100 surface plankton net tows were conducted onboard various research vessels. Sixty-two percent of all tows contained plastics and the largest sample contained 1,069 pieces, which would equal 580,000 pieces/km. Although plastic production increased steadily after the year 2000, it is remarkable that this study showed an increase in the abundance of plastic debris only up to the year 2000, whereas the period from 2000 to 2008 showed barely any increase in plastic debris (Law et al. 2010).

The Agalita Marine Research Foundation is an institution that has sent many expeditions across the North Pacific gyre, and is planning more expeditions to other gyres like the South and North Atlantic. Nevertheless, abundance information on the incidence of floating plastic debris in the ocean is very limited. Gaining insights

into the extent of such floating plastic pollution is almost impossible because of the immense surface area of the oceans. Nevertheless, the few studies that are available have produced enough information to suggest that humanity should be alarmed at the magnitude of floating plastic pollution, and the fact that it has become a serious waste problem.

In addition, as recently shown (Zarfl and Matthies 2010) plastic microdebris fragments, termed microplastics, also occur in oceans worldwide, including even Antarctica, where they are brought by ocean currents.

3.5.2 Plastic Debris Washed Ashore

Plastic debris is very commonly found on many beaches. Much of what is known about the distribution and origin of plastic debris comes from the monitoring of debris that has been stranded on beaches.

Monitoring Beach Debris

Surveys of marine debris accumulation on beaches have been used as the most common way to estimate the load of marine debris at sea, and they can also be used for public education and environmental awareness. Beach areas are easily accessible, and permit low-cost monitoring, although obtaining reliable datasets on beach pollution requires use of the same protocol and sampling methods. Therefore, the United Nations Environmental Programme and the Intergovernmental Oceanographic Commission have developed a standardized marine litter sampling protocol (Cheshire et al. 2009). This protocol includes several important specifications: beach areas to be surveyed should have a slope between 15° and 45° (shallow mudflats are not considered sample areas) and should be from 0.1 to 1 km wide. The beaches should have clear access to the sea and not be blocked by any anthropogenic structures. The surveys should be performed every 3 months throughout a period of 5 years, and the site should not be subjected to any other marine debris collecting activities. The items collected should be categorized into different classes by weight, size, and material type (Cheshire et al. 2009).

Plastic Accumulation on Beaches

Quantities of plastic debris items are highly variable over the course of any 1 year and per location, but numbers of more than 40,000 plastic items (mostly plastic pellets) per m² are not uncommon (Gregory 1978; Thompson et al. 2009a). The accumulation of plastic debris is greater near densely populated areas and on more frequently visited beaches; plastic litter on beaches are primarily sourced from adjoining land areas.

Ross et al. (1991) studied the sources of persistent marine litter in the Halifax Harbour, Canada, and concluded that 62% of the total litter, whereof 54% was plastics, originated from recreation and land-based sources. In contrast, at beaches far from urban areas, most plastic debris consisted of discarded fishing gear and litter. Derraik (2002) reviewed studies on the percentage of plastics in marine debris and concluded that the proportion of plastics varied between 60% and 80% of total marine debris.

A study in Singapore (Ng and Obbard 2006) showed that plastic microdebris accumulated in both seawater and in the sediment of Singapore beaches. The microdebris, containing polyethylene, polypropylene, polystyrene, nylon, polyvinyl alcohol, and acrylonitrile butadiene styrene, were derived from the physical and chemical fragmentation of larger plastic debris. The cleaning of such microscopic items from beaches is almost impossible, and moreover, photo-oxidative degradation of the debris does not occur because it becomes buried beneath beach sediments. In another study, performed along the tropical beaches of Northeast Brazil, the quantity, composition, and distribution of marine debris over a beach area of 150 km south of Salvador city, was examined. It was observed that at some locations the marine debris consisted of 90% plastics and Styrofoam. The average density of the debris was 9.1 items/m² being threefold higher than north of Salvador city, as a result of the southward littoral drift (Santos et al. 2009).

In 2010, the abundance of plastic particles in Belgian coastal waters and beach sediments showed a generally high distribution of microplastics (Claessens et al. 2011). Concentrations up to 390 particles/kg dry sediment were observed. The most abundant particles were plastic fibers (59%) and plastic granules (25%). The study results suggested that fresh water rivers are a potentially important source of microplastics, and showed temporal trends of increased microplastics in coastal sediments.

In a recent study, the effect of small plastic debris on water movement and heat transfer through beach sediments was investigated (Carson et al. 2011). Sediment cores from a beach known for plastic accumulation were compared with a beach where plastics were less common. The great majority (95%) of cores from the former beach contained plastic particles that were concentrated in the top 15 cm of the sediment, which sediment was also coarser grained and more permeable. Artificial cores were constructed that had different plastic-to-sediment ratios, and adding plastic significantly increased sediment permeability. Furthermore, sediments that contained plastics warmed more slowly and reached lower maximum temperatures. These changes can have a serious effect on beach organisms, including those that have temperature-dependent sex-determination, such as sea turtle eggs (Carson et al. 2011).

3.5.3 Plastic Debris on the Seabed

Marine debris is found resting or drifting on the seabed at all depths. It is estimated that in the North Sea up to 70% of marine litter ends up on the seabed.

Monitoring Debris on the Seabed

Data on the abundance of plastic debris in the benthic environments is still very limited, and is restricted by sampling difficulties and the costs of research into deep seabed ecosystems. Therefore, most scientists who have investigated seabed debris have focused their attention on continental shelves (Barnes et al. 2009). Benthic litter can be surveyed by using trawls and camera equipment towed behind a boat, or by direct visual observation by divers. The latter can only be performed in shallow waters, while trawls can also be used to probe deeper parts of the sea. When observations are made with towed equipment, like trawls, great care should be taken by researchers. Such methods can have a huge environmental impact from the by-catch of fish and the physical damage wrought on the benthic environment. A good example of this collaboration is the “Fishing for Litter” program. This program aims to reduce and survey the amount of marine debris by providing fishing boats with large bags for the deposit of marine sourced litter.

Plastics on the Seabed

In the North Sea, study results have indicated that an average of 110 pieces of debris per km² occurs on the seabed. If this number is extrapolated to the whole North Sea, a total of 600,000 m³ of marine debris would be present on the seabed. In the Mediterranean, at a depth of 2,500 m, 300 million pieces of marine debris were found while surveying France and Corsica (UNEP 2001). In Dutch waters, the “Fishing for Litter” project has already collected 500 ton of debris between 2000 and 2006. This debris consisted of truck tires, fridges, large tree trunks, packaging material, lost shiploads, fishing gear, and ropes, among other things (KIMO 2010).

In 2004, the abundance and composition of marine benthic debris was investigated in the eastern Mediterranean on some coastal areas of Greece (Fig. 3). The mean total density of marine debris was estimated to be 15 items/km², ranging from 0 to 251 items/km², with plastics being the dominant form of debris (55.47%) (Katsanevakis and Katsarou 2004). In a second study conducted in the Patras and Echinadhes Gulfs of Western Greece, marine debris from fishing boat trawls was examined. The density of this debris in these two Gulfs was respectively 89 and 240 items per km². Again, the dominating form of debris consisted of plastic items (Stefatos et al. 1999).

3.6 Conclusions

Plastics introduced into the environment end up in different debris pools; floating on the surface, sinking to the seabed or washed ashore (Fig. 4). Floating plastics appear to accumulate in current waters and are very abundant in the world’s gyres. Approximately 70% of all floating plastic objects are believed to eventually

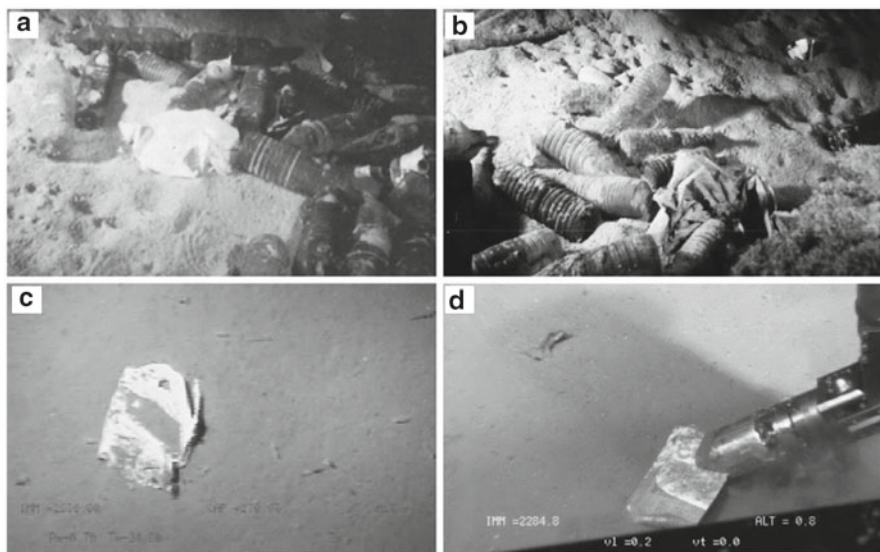


Fig. 3 Accumulation of debris at the seafloor in Mediterranean canyons (**a** and **b**; plastic bottles at 1,000 m depth in the Marseille canyon) and above the polar circle, under an ice sheet (**c** and **d**; plastic bags at 2,200–2,600 m depth at Hausgarten, Fram strait) (reprinted with permission from Barnes et al. 2009)

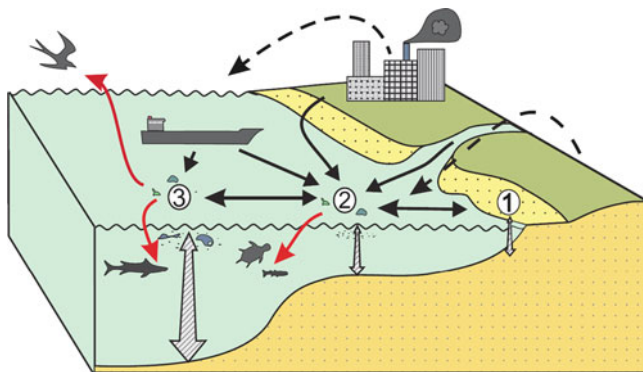


Fig. 4 A schematic diagram showing the main sources and movement pathways for plastics in the marine environment: (1) debris washed ashore on beaches, (2) debris in coastal waters, (3) debris in the open ocean, which may also sink to the seabed. *Dashed arrows* indicate wind-blown debris, *black arrows* waterborne debris and exchange between debris pools, *red arrows* effects on marine life, and *striped-gray arrows* vertical movement through the water column (adapted from Ryan et al. 2009)

sink to the seabed. Near densely populated areas, plastic debris consists primarily of user plastics. In contrast, in areas remote from human activity the debris mostly consists of abandoned, discarded, or lost fishing gear. The fishing industry is responsible for the largest input (50–90%) of total plastic marine debris to the oceans. Therefore, reducing loss and abandonment/discard by the fishing industry

would significantly reduce the input of marine litter, and its effects on marine life. However, the plastic items that are present in the marine environment will fragmentize into smaller particles, microplastics, which are persistent and only slowly degrade. Recent studies showed that these microplastics occur nearly everywhere in the world's oceans including Antarctica.

4 Impact of Plastics on the Marine Environment

4.1 Introduction

The properties that make plastics such desirable materials for modern society can make them lethal for wildlife, when introduced into the environment. Numerous species are affected by plastic pollution, primarily because organisms become entangled in plastic nets, or plastic objects are ingested when organisms mistake plastic debris for food (Laist 1997). Another problem of plastic pollution is that it facilitates the transport of species to other regions; alien species hitchhike on floating debris and invade new ecosystems, thereby causing a shift in species composition or even extinction of other species (Aliani and Molcard 2003). Plastics also transfer contaminants to the environment or to organisms when ingested (Teuten et al. 2009). In addition to impact on marine life, plastic debris can also damage marine industries (entangling propellers and blocking cooling systems). It has been estimated that marine debris damage to the marine industry in the Asia-Pacific region costs \$1.26 billion annually (McIlgorm et al. 2011).

4.2 Mechanical Impact

It was shown that at least 267 marine species worldwide suffer from entanglement and ingestion of plastic debris (Laist 1997). When such contacts occur, organisms are seriously affected in ways that quite often results in death.

4.2.1 Entanglement

It is very difficult to estimate what the total effect of plastic debris in the ocean is, or to predict the consequences for organisms that ingest or otherwise contact that debris, because it cannot be directly observed. By contrast, entanglement can be observed, and is the most visible effect of plastic debris on organisms in the marine environment. Laist (1997) studied and composed a comprehensive list of species that suffered from entanglement and ingestion, and estimated that a total of 136 species are being affected by marine debris entanglement (Table 2). Nevertheless, the exact extent of entanglement faced by marine organisms is difficult to quantify, because entanglement generally occurs in areas remote from human activity.

Table 2 Number and percentage of marine species that have documented entanglement and ingestion records (Reprinted with permission from Laist 1997)

Species group	Total number of species worldwide	Number and percentage of species with entanglement records	Number and percentage of species with ingestion records
<i>Sea turtles</i>	7	6 (86%)	6 (86%)
<i>Seabirds</i>	312	51 (16%)	111 (36%)
Penguins (Sphenisciformes)	16	6 (38%)	1 (6%)
Grebes (Podicipediformes)	19	2 (10%)	0
Albatrosses, Petrels, and shearwaters (Procellariiformes)	99	10 (10%)	62 (63%)
Pelicans, Boobies Gannets, Cormorants, Frigatebirds, and Tropicbirds (Pelicaniformes)	51	11 (22%)	8 (16%)
Shorebirds, Skuas, Gulls, Terns, Auks (Charadriiformes)	122	22 (18%)	40 (33%)
Other birds	–	5	0
<i>Marine mammals</i>	115	32 (28%)	26 (23%)
Baleen Whales (Mysticeti)	10	6 (60%)	2 (20%)
Toothed Whale (Odontoceti)	65	5 (8%)	21 (32%)
Fur Seals and Sea Lions (Otariidae)	14	11 (79%)	1 (7%)
True Seals (Phocidae)	19	8 (42%)	1 (5%)
Manatees and Dugongs (Sirenia)	4	1 (25%)	1 (25%)
Sea Otter (Mustellidae)	1	1 (100%)	0
<i>Fish</i>	–	34	33
<i>Crustaceans</i>	–	8	0
<i>Squid</i>	–	0	1
Species total		136	177

Entanglement can cause death by drowning, suffocation, strangulation, or starvation (Allsopp et al. 2007). Very often, birds, small whale species, and seals drown in ghost nets, lose their ability to catch food, or cannot avoid predators because of their entanglement (Derraik 2002).

Coastal and Marine Birds

Many birds in the marine environment dive for food, and thereby come into contact with plastic debris. The greatest causes of entanglement by seabirds are fishing lines and six-pack rings. Both materials are often transparent and difficult to see. If seen, they can be mistaken for jellyfish and other food (Allsopp et al. 2007).

The gannet is one marine bird species that is endangered by plastic debris. As a “plunge-diver,” the gannet dives from great heights into the ocean and can thereby be caught by ghost nets or other debris. A study at the island of Helgoland in



Fig. 5 A Grey Seal inside a seal shelter at Texel, The Netherlands. The seal was entangled in a nylon thread which had cut into the flesh and damaged the backbone. It suffered from internal bleeding and symptoms of paralysis. Because of its incurable injuries the veterinarian euthanized this animal (De Wolf 2008)

Germany, which hosts a large gannet colony, showed that between 1976 and 1985, 29% of dead gannets found had become entangled in net fragments (Schrey and Vauk 1987). Helgoland is generally a safe habitat for these birds and one of the few threats is entanglement while foraging. Another study, performed in the Netherlands between 1970 and 2000, showed that, of the total number of dead gannets found (1,413), 5.9% (83) had died from entanglement by fishing nets, rope, nylon fibers, nylon line, or other unidentified plastics (Camphuysen 2001). The numbers of entangled gannets have increased over time, which may relate to the increasing amount of plastics produced in Europe. The dead gannets found on the Dutch coast were far away from their colony and were often transmigrating to other places, in contrast to the gannets from Helgoland. There is a chance that a portion of the gannets in the second study died from exhaustion, which may explain the difference in entanglement percentages. Entanglement is probably most common for gannets, albatrosses, a few gull and penguin species, and petrels (Laist 1997).

Seals

Many seal species are curious and playful, and especially young seals are attracted to plastic debris and swim with it or poke their heads through loops. Plastic rings, loops, or lines easily glide onto the seal's neck, but are difficult to remove due to the backward direction of the seal's hair. As the seal grows, the plastic collar tightens and strangles the animal or severs its arteries (Fig. 5). When foraging, many seals become entangled in submerged fishing nets, especially in the North Sea where their vision is limited. After entanglement in these nets the animals are not able to reach the water surface, and drown. Every year fykes⁶ in Dutch coastal waters

⁶A fish trap consisting of a net suspended over a series of hoops, laid horizontally in the water.

causes the death of 15 gray- and harbor-seals, and in 1987, during a search for new feeding grounds, 60,000 harp seals died in stake nets in Norway (De Vleet 2010).

Hanni and Pyle (2000) studied the synthetic-material entanglement of California sea lions, northern elephant seals, steller sea lions, pacific harbor seals, and northern fur seals, between 1976 and 1998, at south-east Farallon Island, California. A total of 914 pinnipeds had indications of entanglement (32%) or displayed constrictions of past entanglement (68%) from various debris types. Most entangled pinnipeds were California sea lions (820), of which 72% had neck constrictions. A total of 68 northern elephant seals were observed to have been entangled primarily by packaging material (59% of the total entanglements) and miscellaneous synthetic materials. Of the 26 entangled Steller sea lions, 15 were observed to have salmon flashers or other hooks hanging from their jaws (Hanni and Pyle 2000).

In a second study performed at the other side of the Pacific Ocean, on the shores of Australia and New Zealand, it was estimated that 1,478 fur seals and sea lions die annually from entanglement (Page et al. 2004). In Australian coastal waters, sea lions were observed to most frequently become entangled with monofilament gillnet, which originated from the shark fishery in that region. In contrast, in New Zealand coastal waters fur seals were observed to primarily become entangled in packaging material, loops, and trawl net fragments that were suspected to originate from regional trawl fisheries (Page et al. 2004).

The material that is responsible for causing entanglement of seals often originates from local fisheries. In many cases, the area where seals forage is also used by humans for shark or trawl fishery. For example, the Farallon Islands are well-known fishing grounds for recreational fishery, and this may have caused the high percentage of flashers embedded in seals of this region.

Whales

Whales also become entangled in marine debris. However, although some whale species are incapable of freeing themselves and consequently drown, the larger size whales often drag fishing gear away with them. This latter type of entanglement can cause strangulation and can affect the feeding ability of the whale in ways that causes starvation (Fig. 6).

In 2005, a study was performed on the entanglement of large whale species in the western North Atlantic Ocean. The purpose of the study was to investigate the entanglement of 31 right whales and 30 humpback whales to determine the types of gear involved. The most common points of gear attachment on the whale's anatomy were the mouth and the tail. Further, 89% of the entanglements were determined to result from pot and gill net gear (Johnson et al. 2005). Pots and gill nets both are located on the seafloor. They are often attached in tandem to each other, and to surface buoys. Large whale species regularly become entangled in these buoy- or connection-lines. According to Johnson et al. (2005), most whale entanglements in the western North Atlantic Ocean involve ground lines. The Provincetown Centre for Coastal Studies, together with several federal agencies, is monitoring the

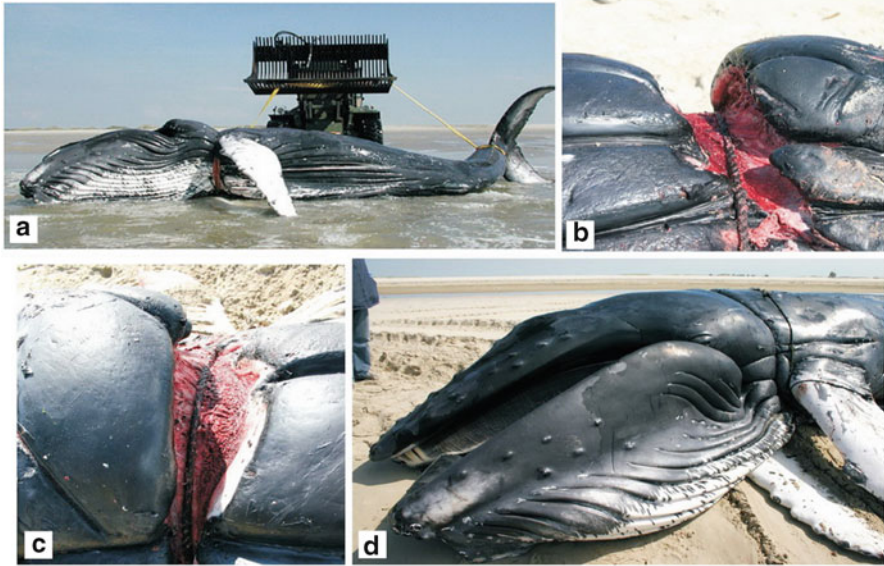


Fig. 6 In June 2004, a Humpback Whale was stranded on the coast of Vlieland an island in the north of the Netherlands. The whale was entangled in a nylon rope that was wrapped around the head. The rope had cut deeply into its body and was probably the cause of the animal's death (**b–d**). The specimen, a young male and approximately 8 m long, was first buried upon discovery by the Dutch Air Force, because it was stranded in a practice area. (**a**) After the photos were shown to experts, the animal was determined to be a Humpback Whale, which is a rare whale species in the North Sea (Bruin 2004)

abundance of whale entanglements in the Atlantic coastal waters of the USA and Canada. Between 1983 and 2009 there were 83 reports of entangled whales in these regions (PCCS 2010).

Fish Species and Ghost Fishing

The incidence of accidental entanglement of fish species is difficult to estimate, because certain fish are “intended” to become entangled in nets. Therefore, research emphasizes by-catch of endangered species. For example, between 1978 and 2000, 28,687 sharks were caught in nets that protected people at popular swimming beaches in KwaZulu, South Africa. Over this period, 53 sharks were found with polypropylene strapping bands around their bodies, and these sharks were evaluated as being significantly underweight (Cliff et al. 2002). Another source of entanglement of fish species is caused by ghost fishing (see Sect. 3.2.1).

Ghost fishing results from fishing gear that continues to function in the water after being discarded or lost (UNEP 2009b). Fishing nets and pots can capture marine organisms, which subsequently die if they cannot escape. In turn, these organisms attract larger predators which also become trapped. When the larger

organisms die they attract smaller scavengers, and so the cycle continues. These fishing nets and pots are death traps for marine organisms, because they do not biodegrade, but rather continue to “fish” for many years (UNEP 2009b).

Sea Turtles

Sea turtles are well-known victims of plastic debris. Juvenile specimens are easily caught in discarded fishing nets, and succumb by drowning. Larger sea turtles are still able to swim with fishing gear attached to their fins or shell, but the debris often affects their ability to feed in ways that eventually results in starvation.

A study on the cause of death among sea turtles stranded at the Canary Islands, Spain, revealed that 70% had died from the influence of human activities, including entanglement by discarded fishing nets (25%). In the same study, it was demonstrated that only 27% of the turtles died from natural causes like diseases (Orós et al. 2005). Plastic debris and other human activities have a big impact on these species worldwide, because six out of seven sea turtle species are known to be affected by entanglement (Table 2) (Laist 1997). Since only 7–13% of the turtles that die from the influence of fishing are washed up on the beaches (Bugoni et al. 2001), studies of stranded turtles alone address only a small part of the total mortality that is caused by fisheries and plastic debris.

4.2.2 Ingestion

Plastic debris that pollutes the marine environment is often ingested by marine birds, mammals, turtles, and fish (Laist 1997). The ingestion of plastics primarily occurs when it is mistaken for food, but can also occur from incidental intake. The ingested material often consists of micro- and meso-debris sized fragments, which sometimes are able to pass through the gut without hurting the organism. In most cases, however, fragments become trapped inside the stomach, throat, or digestive tract and cause damage (e.g., sharp objects) or a false sense of fullness, which will result in starvation.

Coastal and Marine Birds

A high proportion of coastal and marine bird species (36% of the 312 species worldwide) ingest plastic fragments (Laist 1997). Although plastics are mainly ingested by birds because they are mistaken for food, they may also already be present in the gut of their prey, or may be passed from adult to chick by regurgitation feeding. Some species feed selectively on plastic fragments that have a specific shape or color (Moser and Lee 1992). Therefore, plastics ingestion by birds is directly related to their feeding habits and foraging techniques. For example, birds that consume fish (piscivores) are less likely to ingest small plastic fragments than are birds that primarily feed on plankton (planktivore); the latter often confuse plastic pellets with their prey (Derraik 2002). A study on the ingestion of plastic particles by sea birds

in the Subarctic North Pacific Ocean showed a great variation in ingestion of plastics between species within the same area, which confirms the correlation between plastic ingestion and feeding and foraging techniques. Robarts et al. (1995) found 4,417 plastic particles in the gut contents of 1,799 birds, of which 76% consisted of plastic pellets. In comparison with an earlier study in the same area, an increasing frequency of ingested plastic particles was found over time (Robarts et al. 1995).

The Laysan albatross accumulates plastic fragments when collecting food for the feeding of its chicks. These plastics are passed on to the chicks by regurgitation. A total of 251 Laysan albatross chicks from Sand Island, Midway Atoll, were examined, and only six did not contain plastic fragments. Of the 245 chicks that carried ingested plastic, a variety of plastic items were found that included hips and shards of unidentified plastic, Styrofoam, beads, fishing line, buttons, chequers, disposable cigarette lighters, toys, PVC pipe and other PVC fragments, golf tees, dish-washing gloves, magic markers, and caylume light sticks. Most of these items were trapped, and were acting to block the stomach or digestive paths of these birds, rather than to damage their guts; such blockage eventually leads to starvation.

The northern fulmar is a planktivore bird species that is often studied for its ingestion of plastics. In 2006, fulmars obtained from fisheries as by-catch in the Davis Strait between Canada and Greenland, were examined for plastic particles; 36% of the total of 42 birds evaluated contained at least one piece of plastic (Mallory et al. 2006). In general, the number, size, and volume of plastics ingested by fulmars in the north of the North Pacific and the North Atlantic Ocean were lower than in fulmars from the southern parts of these regions. Study results from the North Atlantic Ocean disclosed an incidence of plastic ingestion by fulmars of 79–99% (Moser and Lee 1992; Van Franeker 1985; Van Franeker and Meijboom 2002) and 84–88% in the North Pacific Ocean (Andrady 2003; Robarts et al. 1995). The composition of plastic debris inside the fulmars also varied; in the David Strait 100% of ingested plastic were fragments of discarded plastic products (user plastics), whereas in the North Sea, only 50% consisted of user plastics (Mallory et al. 2006; Van Franeker 1985). Apparently there are regional differences in number, size, and volume of ingested plastics by fulmars, which can be explained by the difference in abundance of plastic debris that occurs near manufacturing centers or areas with intensive shipping traffic. The OSPAR commission, aiming to protect and conserve the North-East Atlantic Ocean, defined acceptable ecological quality as the situation in which no more than 10% of fulmars exceed a critical level of 0.1 g of plastic in the stomach (OSPAR 2008). In a recent study on the abundance of plastics in stomachs of northern fulmars from the North Sea, 1,295 dead beached fulmars were sampled from various European countries, and it was observed that 58% of the birds exceeded the critical level of 0.1 g of plastic; these amounts greatly exceeded the acceptable ecological quality critical level of 10% (Van Franeker et al. 2011).

Seals

Ingestion of plastic fragments is far more commonly reported for birds than for seals. The reason for this may also result from the small sample size prevalent in

seal studies. Feces from fur seals at Macquarie Island, Australia, were examined for plastic fragments in 2003. A total of 164 plastic fragments, mostly polyethylene (93%), were found in the scat of 145 seals, which is more than one fragment per seal. All fragments consisted of user plastics. According to the otoliths, and compared to plastic ingestion by fish in other studies, these fragments were probably not directly ingested by the seals, but rather were accumulated in the fish they consumed (Eriksson and Burton 2003).

Whales

Twenty-eight of 75 whale species, including toothed whales and baleen whales, were reported to have ingested plastic debris (Baird and Hooker 2000; Laist 1997). Most whales that ingest plastic debris live in remote areas and may sink after they die. This, and the fact that most whale species are protected, makes it difficult to study the incidence of plastic ingestion by whales. The sample size is often very small, and is limited to specimens that have been washed ashore. Nevertheless, if one specimen is found to be affected by ingestion of plastic debris, it is probable that other individuals from the same species run comparable risks.

A harbor porpoise, found dead on a beach near Pictou, Canada, died from ingesting a balled up piece of plastic that measured 5 by 7 cm when stretched out. Upon examination, the plastic was found to have blocked the digestive tract, resulting in the accumulation of bones, half digested fish and intact fish in the digestive track. The harbor porpoise had died from starvation (Baird and Hooker 2000). Another report showed that the death of a Sperm whale, which had washed ashore in Texas, USA, had died from ingesting a corn chip bag, plastic sheets, a garbage can liner, and a bread wrapper. In one final example, the death of a beaked whale that washed ashore in Brazil was believed to have resulted from the ingestion of a bundle of plastic threads (Derriak 2002).

Walker and Coe (1989) reported 43 incidents of debris ingestion in 16 stranded toothed whale species. Of these incidents, 80% resulted from plastic debris, mostly plastic bags and sheeting. The authors stated that the ingestion of debris by most toothed whales occurred primarily as incidental ingestion as they were consuming benthic prey. Most reported incidents occurred on the east and west coasts of North America. Variability among these reports may have resulted from regional differences in surveys, recovery, and necropsy, rather than true geographical differences (Walker and Coe 1989).

Data from a study on the ingestion of plastics by Franciscana dolphins in Argentina indicated that 28.1% of the 106 examined dolphins had plastic debris in their stomachs. Most debris (64.3%) consisted of plastic packaging (cellophane, bags, and bands) and a lower proportion (35.7%) consisted of fishing gear fragments. A sharp increase in the occurrence of ingested plastic debris was found in younger dolphins during their weaning phase. Such dolphins may have misidentified what constituted food, or plastic debris, because they had yet to learn what is and is not edible (Denuncio et al. 2011).

Fish

Plastics ingestion by fish has received little attention, with most reports recording only incidental ingestion events. Tiger sharks are known to ingest various items of plastic debris, including plastic bottles, caps, bags, and foil (Randall 1992). Authors of a study performed in the Bristol Channel observed ingested plastic (polystyrene) fragments in the gut of 21% of the flounders examined. Similar fragments were found in 8 of 13 fish species caught along the New England coast, USA (Derraik 2002). Laboratory experiments have proven that some larval and juvenile species of mullet and spot feed on polystyrene fragments. Further, some larval and juvenile fish species in the field were found to have plastic pellets or fragments thereof in their guts. In addition, some adult species had a wide range of material in their guts, from plastic fragments to whole plastic cups. The ingestion rate of plastic particles by mesopelagic fishes at the North Pacific Subtropical Gyre was estimated to be between 12,000 and 24,000 ton/year (Davison and Asch 2011). However, little is known about the impact of plastics ingestion among fish species. This is largely because sampling has not been sufficiently frequent, and there is almost no evidence to determine if ingestion is an important cause of mortality in fish (Hoss and Settle 1990).

Sea Turtles

Sea turtles are among the marine species which are most threatened by plastic debris. Various studies showed that sea turtles do ingest plastic debris (Bugoni et al. 2001; Derraik 2002; Orós et al. 2005). Plastic debris, like bags and sheets, is often transparent and can be mistaken for jellyfish, which is a key diet item for most sea turtles. Furthermore, sea turtles are endangered species, and if plastic intake increases their mortality, the consequences for sea turtle populations around the world may be quite serious.

One turtle found in New York was reported to have ingested 180 m of heavy fishing line (O'Hara et al. 1988). In a study in southern Brazil the contents of the stomach and esophagus of 38 dead stranded Green Turtles was examined. Results were that 60.5% of the green turtles had ingested plastic debris, and this debris caused the death of 13.2% of the green turtles examined. The ingested materials were comprised mostly of plastic bags and white or colorless plastic pieces (Bugoni et al. 2001). Authors of a study in the Mediterranean Sea analyzed debris ingested by 54 juvenile loggerhead turtles. Forty-three of these turtles had ingested marine debris, of which 76% consisted of plastics. Loggerhead turtles are general predators and display little prey discrimination while foraging. This was confirmed by a large variety of plastic items of different colors and shapes found inside their digestive tracts (Tomás et al. 2002). In comparison, green turtles have a selective feeding pattern (Coyne 1994), which was reflected in the more uniform kind of debris found in these animals.

4.3 *Chemical Impact and Ecotoxicology*

Plastics are considered to be biochemically inert; because of their macromolecular structures, they neither react with, nor penetrate the cell membrane of an organism. However, most plastics are not pure. Besides their polymeric structure, they consist of a variety of chemicals that all contribute to a certain property of the plastics they comprise. These chemicals are called additives and they function as described in Sect. 2.3 above. Additives are mostly of small molecular size, are often not chemically bound to a polymer and are, therefore, able to leach from the plastics. Being primarily lipophilic, they penetrate cell membranes, interact biochemically, and cause toxic effects. Moreover, plastic debris in the marine environment not only contains additives, but also contains chemicals (contaminants) adsorbed from the surrounding water. The hydrophobic surface of plastics has an affinity for various hydrophobic contaminants, and these are taken up from the surrounding water and accumulate on, and in the plastic debris. This mechanism receives great attention for microdebris or microplastics, because they are easily ingested by organisms and constitute a pathway for chemicals to enter an organism (Andrady 2011).

In summary, plastic debris in the marine environment can contain two types of possible toxic contaminants: (1) additives and (2) hydrophobic chemicals that become adsorbed from the surrounding water (Teuten et al. 2009).

4.3.1 **Toxic Additives: Phthalates and Bisphenol A**

The release of additives into the environment changes the properties of polymers and affects living organisms. Bisphenol A (BPA) is a constructive monomer that is used in polycarbonate and as a plasticizer, stabilizer, and antioxidant in other plastics such as PVC (Yamamoto and Yasuhara 1999). There are many studies that address the leaching of BPA from polycarbonate or other plastics into the aquatic environment (FDA 2010; Sajiki and Yonekubo 2003; Yamamoto and Yasuhara 1999). Sajiki and Yonekubo (2003) reported that BPA was easily leached from polycarbonate tubes into seawater at 37°C. The leaching rate depended on the temperature of the surrounding water, which can be a concern along tropical seashores in the summertime.

Phthalates are a group of chemicals that are widely used as plasticizers, primarily in PVC polymers. Phthalates and BPA are proven endocrine disruptors. These agents disrupt the functioning of the hormone system, and have received much attention because of their ubiquitous presence in the environment and in humans (Diamanti-Kandarakis 2009; Koch and Calafat 2009; Sax 2009). Phthalates and BPA can leach into the environment, decreasing the flexibility of plastics and affecting reproduction, impairing development, and inducing genetic aberrations in a variety of organisms (Teuten et al. 2009). In a study published in 2009, the effects of phthalates and BPA were examined on several fish, crustacean, and amphibian species; results were that these chemicals affected development and reproduction of a wide range of species. The authors of this study reported alterations in the number of

offspring produced, reduced hatching success, and disruption of larval development of molluscs, crustaceans, and amphibians by low concentrations of BPA and phthalates. Fish species were affected only by relatively high doses of these chemicals, and these demonstrated species-specific sensitivities to these compounds (Oehlmann et al. 2009).

4.3.2 Toxic Additives: Flame Retardants

Flame retardants are also present as additives in plastics and have been added to many common products. The majority of flame retardants are brominated molecules and are referred to as brominated flame retardants (BFRs). BFRs are widely used in plastic products because they affect material properties in only a minor way, and are very effective in preventing ignition. However, they are also present as contaminants almost everywhere in the world's environment; they exist in air, rivers, and waters up to the Arctic regions. BFRs bioaccumulate in the marine food web, including in Canadian Arctic belugas (Tomy et al. 2008) and blue mussels (Gustafsson et al. 1999). Some BFR congeners cause reproductive and carcinogenic effects (Darnerud 2003), disrupt endocrine systems, and cause neurotoxicological effects on mammals and aquatic organisms (Legler 2008).

4.3.3 Adsorption of Contaminants by Plastic Debris

In the marine environment, adsorption of contaminants by polymers is primarily studied with mesoplastic and microplastic debris. Adsorption reduces the transport and diffusion of contaminants. Hydrophobic organic contaminants have a greater affinity for plastics like polyethylene, polypropylene, and PVC, than for natural sediments (Teuten et al. 2009). Polychlorinated biphenyls (PCBs) are a group of organic compounds that once were used as insulating fluids and coolants, as plasticizing and stabilizing additives in PVC, as flame retardants (before the introduction of BFR as a flame retardant), electronic components, and much more. Although PCBs have been banned since 1977 in the USA, and since 1985 in the Netherlands, they have been spread throughout the environment by leakage, dumping, and leaching (EPA 2010), and are present in waters all over the world. Figure 7 shows the PCB concentrations in plastic pellets that were washed ashore. The concentrations in plastics were highest in samples taken along the coasts of the USA, followed by Japan and Europe. Such differences are caused by a differences in PCB usage and production; of the total global PCB production, the USA produced more than half, whereas Africa, Australia, and tropical Asia contributed only minimal amounts (Teuten et al. 2009). In 2001, results of a study on the adsorption of toxicants by plastic pellets along the Japanese coast showed that pellets adsorb PCBs from the surrounding seawater. Virgin polypropylene pellets were used in a 6-day field experiment and increased PCB concentrations were observed throughout the experiment. Moreover, different plastics were observed to have different adsorption capacities; polyethylene pellets adsorbed four times more PCBs than did polypropylene pellets (Mato et al. 2000).

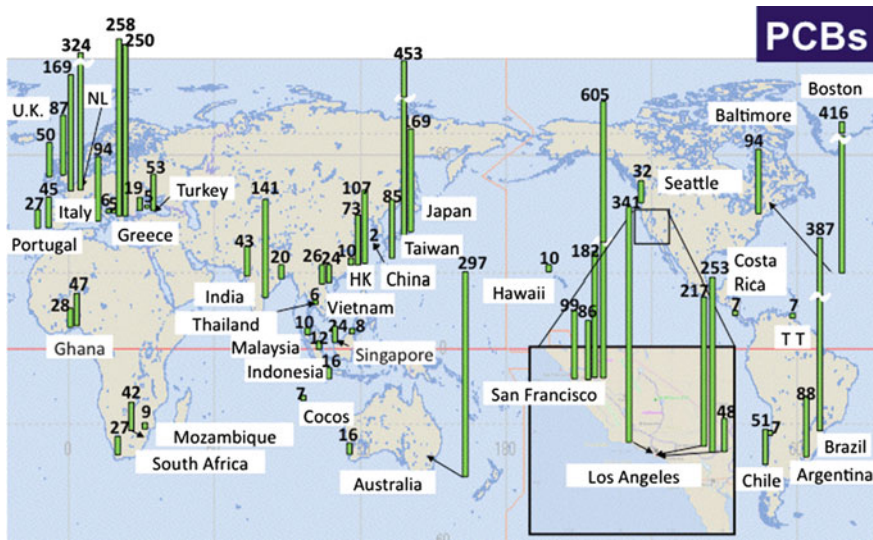


Fig. 7 The concentrations of PCBs that exist in plastic pellets washed ashore. The USA is responsible for half of the world's total PCB production. Therefore, the highest concentrations were found along the US coasts (reprinted with permission from International Pellet Watch 2010)

In addition to PCBs, plastic pellets also adsorb other chemicals, including the pesticides hexachlorocyclohexane (HCH), dichloride diphenyl trichlorethane (DDT) and its metabolites DDE and DDD, and the polycyclic aromatic hydrocarbons (PAHs) that are produced during the burning of fuels. Many of these contaminants are carcinogenic, mutagenic, and/or teratogenic. Adsorption of contaminants can also reduce contaminant biodegradation. Thus, plastics not only adsorb and transport contaminants, but may also increase their environmental persistency (Teuten et al. 2009).

International Pellet Watch (IPW) is a global monitoring program for persistent organic pollutants (POPs). IPW uses plastic resin pellets to monitor the concentrations of contaminants in pellets that are washed ashore. The types and concentrations of chemicals found in these pellets are then used to calculate the concentration of contaminants in the water. This sampling approach is relatively cheap compared to water, sediment, and biological sampling-monitoring approaches and can be used to build maps such as the one that is presented in Fig. 7 (IPW 2010).

4.3.4 Transfer of Contaminants from Plastics to Organisms

Most marine organisms obtain contaminants from plastics by ingesting plastic debris. Adsorbed contaminants can leach into digestive fluids and can be transferred to other tissues. Toxicants may bioaccumulate in the tissues to produce high tissue

toxicant concentrations. Toxicant concentrations may also increase through transfer within a food web (biomagnification). Higher trophic level organisms are exposed to enriched concentrations of contaminants via their prey. However, researchers have shown that some contaminants, like PAHs, do biomagnify less with increasing trophic level (Takeuchi et al. 2009). Notwithstanding, these contaminants are found in marine organisms at high trophic levels (De Laender et al. 2011; Mato et al. 2001).

Results of a study performed in 1988 showed a positive correlation between ingesting plastics and PCB concentration in fat and eggs of 20 female great shearwaters (Ryan et al. 1988). Results from a 2008 feeding experiment proved that PCBs were transferred from contaminated plastics to streaked shearwater chicks. Chicks fed fish laced with polyethylene pellets that were contaminated by PCBs contained PCB residues that were threefold higher than that of the control group (Teuten et al. 2009). These results confirmed that POPs (including PCBs) are transferred to organisms through plastics. However, the authors of a recent study stated that the relative importance of this uptake route is limited compared to other exposure pathways (Gouin et al. 2011). Nevertheless, according to some studies, the ingestion of plastics could play a significant role in the accumulation of contaminants by marine organisms.

In recent years, microplastics have received increasing attention because they are easily ingestible and thereby form a pathway for chemicals to enter organisms, including plankton species (Andrady 2011; Zarfl et al. 2011). As plankton species form the foundation of every food web, any threat to them can have serious effects. The transfer of contaminants within food webs is prevalent everywhere in the marine food web and may even affect nonmarine species such as polar bears (De Laender et al. 2011) and humans (Bocio et al. 2007).

4.4 Use of Plastic Debris by Marine Organisms

Floating natural debris, e.g., trunks from trees or volcanic rocks, have always provided a way for organisms to be transported around the world's oceans. However, because large amounts of plastics have been introduced into the marine environment during the last decades, an increase in marine rafting has been reported. Organisms like algae, mussels, covered with marine organisms have been found floating in the Pacific Ocean, and often wash ashore (Aliani and Molcard 2003; AMRF 2010). Most natural debris is heavy and driven by currents. In contrast, plastic rafts are light weight objects, and are often driven by wind when not totally submerged. Therefore, the species that attach themselves to these plastic items can travel in all directions to colonize new areas. In a study on hitch-hiking of organisms on floating debris, it was reported that an exotic barnacle (*Elminius modestus*) had attached itself to plastic debris found near the Shetland Islands (Barnes and Milner 2005). The incidence of anthropogenic debris more than doubled the rafting opportunities for organisms and is a serious threat to global biodiversity (Barnes 2002).

4.5 Conclusions

Entanglement and ingestion of plastics in plastic debris are the two main causes of mortality in marine organisms. Approximately 267 marine species are known to be affected by entanglement and/or ingestion. The number of affected species may be much higher, since many organisms live in areas remote from human activity. Marine mammals, turtles, and plunge-diving bird species suffer most from entanglement; they get stuck in nets, six-pack rings, or fishing lines and die from starvation, suffocation, or strangulation. For seal species, these harmful plastics often originate from local fisheries that exist in their foraging area. Seals and small whale species and turtles drown from entanglement in (ghost) nets or old fishing gear. The larger whale species drag nets with them, and then suffer from strangulation and starvation as the debris prohibits their ability to catch food.

Marine bird species and turtles are most affected by the ingestion of plastics. They mistake plastics for food and some selectively feed on plastic items. Most marine mammals accumulate plastics in their bodies by feeding on fish that have ingested plastic fragments. There are also cases of ingestion by whales, although the sample size is small for this species (as for other aquatic mammalian species), and often is only based on specimens that have accidentally washed ashore.

In addition to the physical impact of plastics, plastic debris in the marine environment can also leach chemical contaminants into the waters that are absorbed by marine species. Most plastics contain additives such as phthalates, bisphenol A, and BFRs, all of which can leach into the environment. Plastic debris is also known to adsorb contaminants from the surrounding water. Polymers often have an affinity for apolar molecules because they have hydrophobic surfaces. Contaminants leach from the plastics and, when ingested, may cause a variety of toxic effects. Recently, microplastics have received increasing attention because they are easily ingested and form a pathway for contaminants to enter organisms as small as plankton. This causes a threat to the basis of the marine food web and can have serious and far-reaching effects, even on nonmarine species such as humans.

5 Reduction, Prevention, and Clean-up of Plastic Debris in the Marine Environment

5.1 Introduction

Although plastic debris is one of the most widespread forms of marine pollution, it is also among the most soluble of all pollution problems that affect the world's oceans. Notwithstanding, the extent and impact of plastic debris in the marine environment is often underestimated, and therefore the prevention, reduction, and control of plastic debris require much more attention, both from governments and from manufacturers. Because of the nature of the plastic debris problem, a wide variety of approaches and strategies is needed to produce a significantly cleaner and safer marine environment (UNEP 2009a).

5.2 *Prevention*

The plastic debris problem in the marine environment results from the lack of global and regional strategies adequate to prevent the introduction of waste into the environment (UNEP 2009a). Only at the end of the 1960s and early 1970s were the first concerns expressed about accumulating plastic debris and its consequences for wildlife (Kenyon and Kridler 1969; Syrek 1975). Since then a number of countries have taken legislative measures at the national level to regulate the marine litter problem. Most importantly, the cooperation among countries has taken regulatory and preventive measures to an international level.

5.2.1 **Legislation**

MARPOL 73/78

In 1983, a United Nations agency, the International Maritime Organization (IMO), introduced the Marine Pollution (MARPOL) convention, an international protocol to prevent and reduce pollution from ships. The protocol is referred to as MARPOL 73/78, from the fact that the convention was signed in 1973 and the protocol was added in 1978. The protocol has been approved by 169 countries, which together are responsible for 98% of the world's total shipping transport by weight. The protocol consists of several measures attendant preventing pollution in the marine environment by ships. Annex V of MARPOL 73/78 regulates pollution by preventing ships to release garbage, and totally prohibits the disposal of plastics anywhere into the sea. Further, it obligates governments to keep terminal facilities and harbors clean of garbage. According to the terms of this agreement, every ship having a weight over 400 t and able to carry more than 14 persons is obligated to maintain a Garbage Record Book, in which records of all disposal operations will be kept. Information required includes the date, time, position of the ship, and description and estimated amounts of garbage that is incinerated or discharged. In addition to maintaining a Garbage Record Book, mariners are asked to prepare a Garbage Management Plan that gives procedures for collecting, storing, and processing onboard waste (IMO 2010).

The Regional Seas Programme

In 1974, the United Nations Environmental Programme (UNEP) initiated the Regional Seas Programme, which aimed to address the accelerating degradation of the world's oceans and coastal zones. The program seeks to create sustainable management and use of the marine and coastal environments by engaging involved countries and creating a plan of action. All Action Plans have a similar approach, but are shaped by each government according to their own needs and environmental challenges.

Today, the program covers 18 coastal and sea areas and has more than 140 participating countries (UNEP 2010). Nevertheless, the legislation is still widely ignored and it is estimated that ships dump 6.5 million tons of plastic into the world's oceans every year (UNEP 2009b). This flagrant disregard of the dumping rules questions whether this regulatory approach is adequate to deal with such a problem. Although this program may help over the long term, the current continuing extent of the plastic dumping problem demands drastic changes in mankind's behavior.

The Marine Strategy Framework Directive

The Marine Strategy Framework Directive (MSFD) introduced in Europe in July 2008 aims at achieving or maintaining a good environmental status (GES) by 2020 (MSFD 2011). This means that EU member states must develop action plans and activities to achieve this "GES." This includes a legislative framework that allows for managing human activities that have an impact on the marine environment, and also integrating concepts of environmental protection and sustainable use. The criteria and methodological standards on GES of marine waters have been set up by the MSFD and are based on existing obligations and developments within the EU legislation. However, some criteria are fully developed and operational while others require further refinement. Therefore, more scientific knowledge on the marine environment is required to develop a better understanding and achieve the Directive's goal (Zarfl et al. 2011).

5.2.2 Alternatives for Plastics

Another way to prevent the input of persistent plastics into the marine environment is to introduce biodegradable plastics. Biodegradable plastics are made of renewable sources, and consist of polymers that are capable of undergoing decomposition into carbon dioxide, water, methane, inorganic compounds, or biomass. Biodegradation of these polymers is achieved by the use of microorganisms that have the ability to catabolize these polymers into less environmentally harmful material (BioPlastics24 2010). The residue of degraded polymers is often used as plant fertilizer and these plants can serve as a new source for manufacturing biodegradable polymers. Recently, progress has been made in developing biodegradable plastics that possess characteristics similar to those of oil-based polymers (Song et al. 2009). Biodegradable plastics, or bioplastics, often have inferior performance compared to traditional plastics because they eventually become permeable to water. Therefore, bioplastic materials are used as disposable items, such as packaging material. The biodegradable polymers that are used are of diverse types. Bioplastics that are based on polylactic acid (PLA) and Plastarch material (PSM) are two of the most commonly used ones in current commercial practice.

Polylactic Acid

PLA is made from starch-rich substances like maize, wheat, or sugar. The bioplastic made from PLA is biodegradable and can, under ideal composting conditions, degrade in less than 60 days. PLA was discovered in 1890, but has only recently entered the market as a biodegradable plastic. Today, PLA is still more expensive to produce than are many traditional plastics, but the price is decreasing as the demand for bioplastics increases (BioPlastics24 2010).

Plastarch Material

PSM is a thermoplastic polymer composed of starch, from corn, that is combined with other biodegradable materials. PSM is one of the few plastics that can withstand high temperatures (up to 125°C). Apart from the fact that it is biodegradable, the material has similar characteristics to those of polyethylene. After serving its useful life, PSM can be incinerated to produce both a nontoxic smoke and a residue that can be used as a plant fertilizer (BioPlastics24 2010).

Bioplastics are renewable and are easily degradable. Although they have existed for as long as traditional oil-based plastics, the market for them is now expanding as a direct result of the high price of oil. There are only a few producers of bioplastic products. NatureWorks LLC is the largest producer of PLA in the world. They use corn to create PLA food packaging, bottles, and shirts. The Indian company Earthsoul uses the biodegradable polymer Master-Bi to produce various products, although they are focused primarily on products for agriculture. In 2002, the US Department of Agriculture (USDA) found a way to use animal waste for bioplastic production. They used the protein in chicken feathers from poultry production as a building block to make plastic. These feather-derived plastics have high strength and are fully biodegradable (USDA 2009). Sony is one of the giants of electronic production that uses NatureWorks' PLA plastic for their famous Walkman®; moreover, the packaging for their Playstation is made from extendable polystyrene, which is recycled from orange peels (JapanFS 2009). Another company, NEC Electronics, has produced a biodegradable mobile phone, which will biodegrade if buried in soil, and importantly, it does not form toxic gasses when burnt. NEC electronics is also developing a biodegradable laptop computer casing that utilizes PLA, with fibers added to improve strength and heat resistance (Bio-Plastic 2009).

5.3 Recycling

Recycled polymeric materials can be reused, which saves production energy and prevents the dumping of materials into the environment. During the last decade, the mechanical recycling industries have showed an encouraging trend, i.e., a 7% annual growth in western Europe (Thompson et al. 2009a). Unfortunately, the recycling rate varies regionally and globally, and only a small percentage of total plastic waste is

Table 3 Plastics production, recovery and disposal in the USA in 2005 (thousands of metric tons) for polyethylene terephthalate (PET), high-density polyethylene (HDPE), polyvinyl chloride (PVC), low-density polyethylene (LDPE), polypropylene (PP), polystyrene (PS), and other plastics

Plastic type	Generation of plastics in municipal solid waste	Recovery	Discards
PET	2,600	491	2,109
HDPE	53.55	473	4,882
PVC	1,491	0	1,491
LDPE	5,864	173	5,691
PP	3,636	9	3,681
PS	2,355	0	2,355
Other	4,982	355	4,981
Total	26,282	1,500	24,782

Data show that only a small proportion of plastics is being recycled. Plastic material from construction and agricultural sectors are not included (Reprinted with permission from Barnes et al. (2009))

currently being recycled (Table 3). In most countries, the form of plastic that are recycled is largely limited to bottles and drink containers (Barnes et al. 2009). Most consumers are keen to recycle, and support for recycling is often very high in most western countries. However, the difference in symbols (SPI Resin Identification Code) printed on different forms of plastic to describe recyclability of the object vary considerably among countries or regions, and is often an obstacle to convenient recycling. This is why, in most countries, all kinds of plastic waste is collected together and is sorted at special stations before being recycled.

Plastic waste often consists of a mixture of different types of plastics, which makes it difficult for recyclers to work with; this problem is caused, in part, because manufacturers and recyclers neither communicate, nor make agreements. The recycling of plastic items is therefore more difficult than the recycling of paper or glass (i.e., three types only—transparent, green, or brown). For example, plastic drinking bottles may consist of a HDPE body, fitted with a polypropylene cap and a steel ring. The variation of forms or components that compose plastic items can be limitless. Therefore, most recyclers collect all kinds of plastics together, melt it down or grind it up and turn it into a new plastic product.

Tie-Tek LLC is a company that produces railroad ties from vehicle tires, plastic bottles, and plastic bags. One mile of railroad made from these ties (3,300 ties) is composed of the equivalent of nine million plastic bags, two million plastic bottles, and 10,000 vehicle tires. Agri-Plas is another recycling company that collects agricultural plastics and turns them into new plastic items for use in agricultural; hence, the plastics from this company form a circle of production and recycling that is continuous.

5.4 Clean-up

Efforts to render new plastics more environmentally friendly, or legislation to reduce persistent polymer input into the environment do not address the burdens of plastic

debris that are already present in our oceans. The clean-up of existing marine debris often falls to local authorities, nongovernmental organizations, and to volunteers. Clean-up costs can be very high, and great efforts are required to motivate a sufficient number of people to assist in clean-up efforts. For example, the Korean government recently removed derelict fishing gear from the deep seabed of the East Sea by bottom trawling with heavy hooks (50–80 kg) and ropes. A total of 207.8 and 252.2 ton of marine debris was removed from the seabed in 2009 and 2010, respectively; most of the debris was comprised of derelict fishing gear. The total cost of this 2-year project was \$ US2.3 million. The use of bottom trawls is dangerous because they are performed by fishing vessels during closed seasons, when the weather is often stormy and typhoons occur. Such clean-up projects have already led to the loss of one ship and five crew members in 2009 (Cho 2011).

There are many projects that aim to prevent, control, or clean-up marine debris. In addition to debris clean-up, most projects also endeavor to educate the community on the importance of reducing marine pollution. Such education includes distributing brochures or giving lectures at local schools. The effort to educate school age children is important because it instills good habits, and establishes a basis for these children to spread their knowledge to others. In addition, there are some projects that go further, by organizing local or general clean-up of marine debris.

One of the largest organizations in Europe that has an international scope, and deals with marine pollution is Kommunenes Internasjonale Miljøorganisasjon (KIMO). KIMO has the aim of contributing to a steady reduction of marine pollution in Europe's seas. One of their projects is called "Fishing for Litter." This project provides fishing boats with large bags for use in the disposal of marine-sourced debris. When full, these bags are collected for disposal. The Fishing for Litter project has successfully removed debris from the sea and has reduced the volume of debris that is washed ashore. Another environmental program is called Clean Up the World. Clean Up the World is held in conjunction with UNEP, and mobilizes 35 million volunteers from 120 countries to positively improve local environments. They organize activities such as the clean-up of coastal areas, education campaigns for local populations and tree planting. The organization Provincetown Centre for Coastal Studies (PCCS) monitors the abundance of whale entanglements in the Atlantic coastal waters of the USA and Canada. In addition to monitoring programs, the organization is also focused on the removal of entangling material from whales.

5.5 Conclusions

The most effective and efficient response to the plastic debris problem in the marine environment is to ban the input of plastics into the oceans. Therefore, several different prevention measures have been implemented. These include (1) legislation that obligates consumers to pay attention to the waste they generate and (2) the introduction and use of alternatives such as biodegradable plastics. Recycling is another option to reduce input of plastics to the marine environment. It not only prevents the

discard of plastics, but also saves material and energy. Removal of the current bulk of plastic debris that is present in the oceans is also needed. Many environmental organizations contribute to this, or have produced action plans to clean beaches and other coastal areas of plastic debris. These organizations also are capable of contributing to the education of communities by drawing inhabitant's attention to the plight marine species face as a result of plastic debris. Education is particularly important, because it is the basis for teaching the next generation to be aware of and address the consequences of discarding plastics and other debris into the world's oceans.

6 Summary

Plastics are cheap, strong, and durable and offer considerable benefits to humanity. They potentially can enhance the benefits that both medical and scientific technology will bestow to humankind. However, it has now been several decades since the use of plastics exploded, and we have evidence that our current approach to production, use, transport, and disposal of plastic materials has caused, and is still causing serious effects on wildlife, and is not sustainable.

Because of frequent inappropriate waste management practices, or irresponsible human behavior, large masses of plastic items have been released into the environment, and thereby have entered the world's oceans. Moreover, this process continues, and in some places is even increasing. Most plastic debris that now exists in the marine environment originated from ocean-based sources such as the fishing industry. Plastics accumulate in coastal areas, at the ocean surface and on the seabed. Because 70% of all plastics are known to eventually sink, it is suspected that ever increasing amounts of plastic items are accumulating in seabed sediments. Plastics do not biodegrade, although, under the influence of solar UV radiation, plastics do degrade and fragment into small particles, termed microplastics. Our oceans eventually serve as a sink for these small plastic particles and in one estimate, it is thought that 200,000 microplastics per km² of the ocean's surface commonly exist.

The impact of plastic debris has been studied since the beginning of the 1960s. To date, more than 267 species in the marine environment are known to have been affected by plastic entanglement or ingestion. Marine mammals are among those species that are most affected by entanglement in plastic debris. By contrast, marine birds suffer the most from ingestion of plastics. Organisms can also be seriously affected from contact with plastics-associated contaminants. Such contaminants are absorbed by floating plastic debris, or the contaminants may derive from plastic additives that are leached to the environment. Recent studies emphasize the important role of microplastics as they are easily ingestible by small organisms, such as plankton species, and form a pathway for contaminants to enter the food web. Contaminants leached from plastics tend to bioaccumulate in those organisms that absorb them, and chemical concentrations are often higher at higher trophic levels. This causes a threat to the basis of every food web and can have serious and far-reaching effects, even on nonmarine species such as polar bears

and humans, who consume marine-grown food. Therefore, resolving the plastic debris problem is important to human kind for two reasons: we are both creator, and victim of the plastic pollution problem.

Solutions to the plastic debris problem can only be achieved through a combination of actions. Such actions include the following: Legislation against marine pollution by plastics must be enforced, recycling must be accentuated, alternatives (biodegradable) to current plastic products must be found, and clean-up of debris must proceed, if the marine plastic pollution problem is to eventually be resolved. Governments cannot accomplish this task on their own, and will need help and initiative from the public. Moreover, resolving this long-standing problem will require time, money, and energy from many individuals now living and those of future generations, if a safer and cleaner marine environment is to be achieved.

Acknowledgements We would like to thank Salko de Wolf and Dirk Bruin for providing some very useful photos.

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Mercury Pollution in Malaysia

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1 Introduction

Mercury is a hazardous pollutant; concern for its environmental presence arises from the human health effects caused by methylmercury through consumption of fresh water and marine fish (Clarkson 1995). Researchers first became concerned about the harmful effects of mercury when anthropogenic sources were released into the marine environment, and caused poisoning episodes (e.g., neurological disorders) in Japan (Minamata and Niigata) (Keckes and Miettinen 1972). This first known human poisoning by mercury from ingestion of seafood occurred in Japan

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between 1953 and 1961. During that period, more than 100 people were affected by eating shellfish, crabs, and fish from Minamata Bay, Kyushu, Japan. The victims developed many serious neurological disturbances, and severe cases produced stupor, coma, exhibiting involuntary movements, tremors, agitation, and convulsions (Deocadiz et al. 1999).

The World Health Organization (WHO 1976) cited three primary potential sources of mercury exposure: mercury vapor in ambient air; mercury in drinking water; and mercury in the diet. The first two sources of exposure are regarded to be minor contributors to human mercury intake. The primary mode of intake of Hg by the general population is by eating carnivorous fish (e.g., tuna, swordfish, halibut, and shark) and marine mammals (e.g., whales, seals) that have Hg residues in their bodies, or from the release of elemental mercury from dental amalgams that may dissolve in saliva and be ingested (Sallsten et al. 1996; ATSDR 2003). People eating locally contaminated fish, or those that have unusually high consumption rates of large carnivorous fish, eventually develop blood levels of mercury that could trigger poisoning symptoms similar to those that occurred in the Japanese outbreak. The source of the mercury poisoning in Japan originated from the industrial release of mercury into a sheltered ocean Bay (Minamata Bay) and into a river (Agano River) (Deocadiz et al. 1999). Occupational exposure is the principal health hazard associated with mercury poisoning (Rowland et al. 1994). For example, most people who become exposed to elemental mercury vapor are those who are employed in mining and chloralkali plants, and in instruments manufacturing plants that use mercury (e.g., laboratory instruments, accumulators, etc.) in their production processes.

Mercury is among the most important of heavy metals that contaminate the environment (Slemr et al. 1985). Recent estimates (Mason et al. 1994) of the global mercury budget indicate that ca. 6,000 and 10,800 ton of mercury are currently present in the troposphere, and in the earth's water bodies, respectively. Although Malaysia has been one of the less polluted urban environments in Asia (ADB 1997), its goal of achieving industrial country status by the year 2020, and its associated rapid economic growth, have started to impose costs from industrial pollution and urban environmental degradation. Among those recent environmental degradation events have been depletion of fisheries, deforestation, pollution of inland and marine waters, soil and coastal erosion, air and water pollution, and increased contamination by industrial wastes (Afroz et al. 2003; WWF-M 2001). Increasing industrialization and urbanization in the ASEAN (The Association of Southeast Asian Nations) region has caused increased mercury inputs into the marine environment (Deocadiz et al. 1999). Heavy metals are major pollutants in Malaysian waterways, and derive predominantly from industrial point source outfalls and from mining activities (Abdullah 1995). There have been numerous research papers published that address the sources of release and contamination by mercury in the Malaysian environment or in its commodities; such contamination, for example, has been found in water bodies, in biota, and in the human body. In this chapter, it is our aim to review and summarize the content of those studies, and to assess the significance of the amounts of mercury found in different Malaysian environmental compartments, and finally, to identify appropriate resolutions.

2 Mercury: Uses and Source of Input into the Environment

Mercury is released into the atmosphere from a variety of natural (Fitzgerald 1986; Xiao et al. 1991; Mason et al. 1994; Lindberg et al. 1995) and anthropogenic sources (Lindqvist et al. 1991; Ferrara et al. 1992; Pirrone et al. 1996; Carpi and Lindberg 1997; Lacerda 1997). Natural sources include volcanoes, soils, forests, lakes, and open oceans (ca. 2,000 ton/year total; Mason et al. 1994). Anthropogenic sources mainly result from combustion processes and waste incineration (ca. 4,000 ton/year total; Porcella et al. 1997). Elevated levels of mercury exist in waters that are remote from anthropogenic emission sources, which indicates that atmospheric deposition is also an important source of contamination (Swain et al. 1992; Rasmussen 1994; Sorensen et al. 1994). Although it is difficult to identify atmospheric deposition sources in remote regions, it is generally accepted that anthropogenic-based emissions have greatly increased relative to natural sources since the start of industrialization (Fitzgerald et al. 1998; Hanisch 1998).

Malaysia is a coastal state whose shores are washed by the Straits of Malacca, along the west coast of Peninsular Malaysia and the South China Sea, along the east coast of the Peninsula and the coasts of Sabah and Sarawak. The Department of Environment (DOE) of Malaysia regularly monitors water and air quality. Malaysian coastal waters have frequently been observed to contain significant levels of mercury. Based on DOE marine water quality monitoring results from 205 coastal-water monitoring stations, mercury has exceeded the interim standard of 0.001 mg/L every year since 1996 (DOE 1996, 1998, 1999, 2000, 2001, 2002, 2003, 2004, 2005, 2006, 2007, 2008). There was, however, fluctuation in the number of samples that exceeded the interim standard for mercury from 1997 to 2008 (Fig. 1). The highest exceedance rate was 18.2% of samples in 2006. Globally, 70% of pollution in the seas is estimated to originate from land-based sources (UNEP 1990).

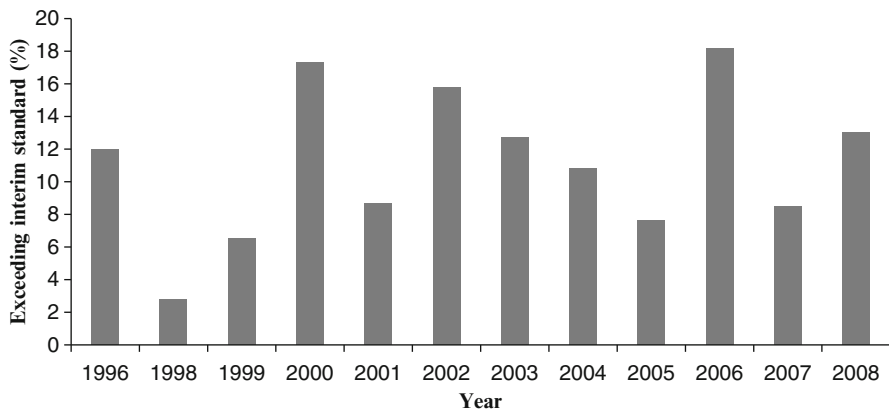


Fig. 1 The annual incidence of mercury residue exceedances in the marine environment of the Malaysian standard (1996–2008) Source: (DOE 1996–2008)

Urbanization, increases in population density, and the intensification of agricultural activities are among the main causes of increasing water pollution (DOE 2008). Significant land-based sources of pollution in Malaysia include agricultural and industrial activities, as well as urbanization concomitant with the rapid pace of industrialization, and the associated increasing amounts of toxic and hazardous wastes generated by a wide range of industrial activities (DOE 2008).

The Straits of Malacca is subjected to a great variety of pollutants due to its strategic location as a major international shipping lane, and the concentration of its agriculture, industry, and urbanization, which predominate on the west Peninsular Malaysian coast. Therefore, the Straits has been a main repository for agricultural, industrial, and domestic wastes originating from land-based activities, whereas shipping activities from operational or accidental discharges have contributed to the pollution of the Straits (Abdullah et al. 1999). The primary concern for the pollution status of the west Peninsular Malaysian coast stems from the fact that discharges reach it from rivers that drain highly the industrialized, heavily tilled, and densely populated areas of the country (Impak 1998). Therefore, the impact of marine pollution in Malaysia, especially mercury, is felt most in the estuarine and inshore coastal areas of the Straits of Malacca. This partially results from the enclosed character of the narrow sea that forms the Straits, and which drain the effluents of several rivers. Land-based developments settled earlier also exist along the west Peninsular Malaysian coast, and the population density of this area is concurrently grown (USM 1976).

The waters of the South China Sea that borders the Malaysian coastline have been relatively free of marine pollution, because development pressures in the coastal states of this region have been rather slack prior to 1970 (USM 1976). Nevertheless, consistent with the economic policies adopted by the Malaysian government to redress poverty, particularly in the less developed coastal states bordering the South China Sea, a concomitant increase in coastal marine pollution has been observed (Hajeb et al. 2009).

Non-natural pollutant sources in the marine environment are derived mainly from manifold human activities, and such activities are not confined to the Malaysian territorial limits. Some pollutants are introduced to these marine waters through atmospheric and aquatic drift inputs that result from activities that occur elsewhere on the earth. Although there has been no significant source of industrial mercury release into the coastal Malaysian environment, such as a chloralkali plant, awareness of the danger posed by mercury pollution has been growing because of the recent rapid pace of industrialization along the west coast of Peninsular Malaysia. According to the Malaysian Industrial Developmental Agency (2007), more metal-related industries have begun operations in the region since the year 2005. Such manufacturing industries include the following: electrical appliances (lamps), control instruments (thermometers), laboratory apparatus, dental amalgams, and raw materials for various mercury compounds such as fungicides, antiseptics, preservatives, pharmaceuticals, electrodes, and reagents. From the environmental point of view, this industrial growth could lead to an undesirable release of metals like mercury into our coastal environment. Metallurgical industries, particularly those involved

in metal plating and galvanizing, are known to release heavy metals such as mercury into their effluents. Mercury has also been found in the effluents of electrical industries (Turney 1971). The pulp and paper industry is known to produce wastes such as sulfites from wood digestion, and chlorinated phenolic compounds from pulp and paper bleaching activities. Mercury, which was formerly used as a slimicide in the paper-making industry, has also been found to exist in paper mill waste effluents (Landner 1978).

The research conducted by Kathirvale et al (2003) on municipal solid waste (MSW) characteristics for the city of Kuala Lumpur showed that the mercury content of MSW was 0.27 mg/kg. Based on this study, the average amount of MSW generated in Malaysia was calculated to be 0.5–0.8 kg/person/day, and in major cities 1.7 kg/person/day. Based on the Environmental Quality Report, a total of 1,302,898.77 metric tons of scheduled wastes were generated in 2008, as compared to 1,138,839.49 metric tons in the year 2007. The mercury content in the wastes produced in 2007 and 2008 was 0.3% and 0.04%, respectively. The main categories of waste produced in Malaysia were gypsum, dross/slag/clinker, oil and hydrocarbon, heavy metal sludge, mineral sludge, and e-waste (DOE 2007, 2008).

3 Mercury Pollution in the Aquatic System

3.1 Rivers and Sea Water

Mercury pollution has occurred in the major rivers of different states of Peninsular Malaysia since 1985. Significant levels have particularly been found in rivers of the more urbanized and industrialized western regions (Table 1). The DOE uses a standard method to analyze for mercury residues annually in river water. Following is an accounting of the Hg residues detected in some of the more urbanized regions of Malaysia:

- In the Merbok river, Kedah, the residues levels reported were, respectively, 0.002–0.46, 0.001–0.005, <0.001, and 0.020 mg/L for the years 1985, 1987, 1989, and 1991 (DOE 1986, 1987, 1989, 1991a, b).
- In the Perai River, Penang, the reported residues were 0.001–0.03, 0.001–0.004, 0.001, and 0.006–0.015 mg/L, respectively, for the years 1985, 1987, 1989, and 1991 (DOE 1986, 1987, 1989, 1991a, b).
- In the Perak River, Perak, the reported residues were 0.001–0.02, 0.001, 0.001, and 0.001–0.05 mg/L, respectively, for the years 1985, 1987, 1989, and 1991 (DOE 1986, 1987, 1989, 1991a, b).
- In the Kelang River, Selangor, the residues reported were 0.001–0.005, 0.001–0.005, 0.001–0.005, 0.001–0.007 mg/L, respectively, for the years 1985, 1987, 1989, and 1991 (DOE 1986, 1987, 1989, 1991a, b).
- Finally, the mercury residues in the Batu Pahat River, Johor, were reported as being <0.001 mg/L in both 1987 and again in 1991 (DOE 1987, 1991a, b).

Table 1 Mercury levels (mg/L) found in water samples from the major rivers of Peninsular Malaysia

Location	Year	Mercury level (mg/L)	Reference
Merbok River	1986	0.002–0.46	DOE (1986)
Kedah	1987	<0.001–0.005	DOE (1987)
	1989	<0.001	DOE (1989)
	1991	0.02	DOE (1991a, b)
Perai River	1985	0.001–0.03	DOE (1985)
Penang	1987	<0.001–0.004	DOE (1987)
	1989	<0.001	DOE (1989)
	1991	0.006–0.015	DOE (1991a, b)
Perak River	1985	0.001–0.02	DOE (1985)
Perak	1987	<0.001	DOE (1987)
	1989	<0.001	DOE (1989)
	1991	0.001–0.05	DOE (1991a, b)
Kelang River	1985	<0.001–0.005	DOE (1985)
Selangor	1987	<0.001–0.005	DOE (1987)
	1989	<0.001–0.002	DOE (1989)
	1991	0.001–0.007	DOE (1991a, b)
Batu Pahat River	1985	–	DOE (1985)
Johor	1987	<0.001	DOE (1987)
	1989	–	DOE (1989)
	1991	0.001	DOE (1991a, b)

DOE's water quality monitoring program for rivers in 1996 resulted in collecting 909 samples in 116 rivers (DOE 1996). Analyses of these samples resulted in classifying thirteen rivers as being polluted; eight of these rivers are located on the west Peninsular Malaysian coast. The mercury levels found in some rivers exceeded the guideline limit (0.001 mg/L), established in the National Guidelines for Raw Drinking Water Quality (2000). Unfortunately, there are no currently published data on the extent of mercury pollution in the sources of those waters.

Mercury levels from samples taken in the Langat River during a 6-month sampling period (September 1984 to February 1985) were in the range of 0.002–0.004 mg/L (Sarmani 1985). The Langat River basin, located south of Kuala Lumpur, is the major source of drinking water for the Kuala Lumpur area, and the water quality from this resource is threatened by the advancing development taking place in the basin. The monitoring of physical, chemical, and biological indicators of water quality commenced for this river approximately 25 years ago (Sarmani 1985). In the late 1980s, Sarmani (1989) demonstrated very low mercury levels (0.002–0.004 mg/L) in the river water of this basin. Total mercury levels in samples collected from the Kelang estuarine waters were in a range of 0.10–6.50 µg/L; these levels were much higher than those detected in the Straits of Malacca and in the South China Sea (Law and Singh 1987).

The levels of inorganic and total mercury were also determined in various river and sea water samples collected from Malaysia by Sakamoto et al. (2004). These authors reported 1.4–41.0 and 1.6–52.0 ng/L of inorganic and total mercury levels in river water samples from the Straits of Malacca and the South China Sea, and

1.0–2.6 and 1.4–2.9 ng/L of inorganic and total mercury in sea water samples from the East China Sea, respectively. The level of mercury in river water was far higher than in sea water, probably because of the contributions from human activities, industry, and agricultural inputs to the rivers. Based on the studies that have been published on mercury pollution in rivers and sea water, most pollution appeared in west coastal waters, and the major chemical form of mercury found in these coastal sea water samples was inorganic mercury.

3.2 Sediments

Among the heavy metals, mercury deserves particular attention because of its highly toxic nature and tendency to biomagnify through the food chain (Zhou and Wong 2000). Sediments were shown to be not only a sink for heavy metals (Salomons et al. 1987), but also to act as a secondary source of metals in the marine environment (Sin et al. 2001). Many studies on mercury in sediments (Chongrak 1982; Krishnakumar and Pillai 1990; Larcercda et al. 1993) have shown that anthropogenic activities are linked to mercury contamination. The high level of mercury found in sediment samples may directly or indirectly reflect the input from anthropogenic activities, such as industrialization, urbanization, and mining (Larcercda et al. 1993).

The total mercury residue levels in wet sediment samples from the Kelang estuary were 0.03–0.40 mg/kg (Law and Singh 1987). Those results suggest that the Kelang estuary carries some degree of mercury pollution. The mercury content in sediment taken from the coastal areas of Kuala Terengganu showed levels of 61 ± 47 , 0.038 ± 0.02 , and 4.81 ± 5.73 ng/g total mercury, methylmercury, and inorganic mercury, respectively. The proportion of methylmercury that existed as part of the total mercury content was 0.02–0.7%, which is rather low for this area (Kannan and Falandysz 1998).

In a comprehensive study performed between 1999 and 2000, total mercury levels in surface sediments of the intertidal area along the west coast of Peninsular Malaysia were determined (Yap et al. 2003). Total mercury levels in these sediments ranged from 3 to 201 $\mu\text{g}/\text{kg}$ dry wt. Compared to the regional data and sediment quality guidelines, the mercury contamination that existed in the intertidal area along the west coast was not serious, except for a few sites that contained anthropogenic-sourced mercury in the collected samples. Low mercury residues were also recorded in sediments collected from recreational sandy beaches, such as Pantai Telok Batek (2.90 $\mu\text{g}/\text{kg}$), Pantai Pasir Bogak (3.84 $\mu\text{g}/\text{kg}$), Pantai Telok Kemang (3.46 $\mu\text{g}/\text{kg}$), Pulau Indah (9.16 $\mu\text{g}/\text{kg}$), Pantai Remis (7.75 $\mu\text{g}/\text{kg}$), and from a remote site at Kuala Muda (6.00 $\mu\text{g}/\text{kg}$). The highest mercury level was recorded in the intertidal sediment from Kuala Juru (201 $\mu\text{g}/\text{kg}$), followed by Jelutong (135 $\mu\text{g}/\text{kg}$) and Bukit Tambun (103 $\mu\text{g}/\text{kg}$) in the state of Penang. The mercury levels in these samples were 35–70 times higher than those found in samples from the recreational sandy beaches. The high mercury contamination from these areas may be due to their proximity to the Prai Industrial Estate, which is an industrial area (Yap et al. 2003).

Mercury in sediment samples from four locations near the Straits of Johore (Pantai Lido, Gelang Patah, Kg. Pasir Puteh, and Tg. Kupang) had levels that ranged from 49.0 to 108 $\mu\text{g}/\text{kg}$.

Studies on mercury residues in sediments were also reported from sites along different coastlines and rivers in Malaysia (Tan 2007; Sakamoto et al. 1999, 2004; Law and Singh 1987). The sediment quality of the Sungai Linggi River basin at Negeri Sembilan and its tributaries (rivers of Linggi, Batang Penar, Paroi, Temiang, Senawang, Kepayong, Kayu Ara, upper Pedas, lower Pedas, Chebong, Siput, Rembau, upper Simin) displayed a wide range of mercury concentrations, 0.31–14.27 $\mu\text{g}/\text{g}$ (Tan 2007). This river drains into the Straits of Malacca at the west Peninsular Malaysian coast, and flows to areas that are near a popular recreational beach (Port Dickson). The study by Tan (2007) indicated that mercury was a major contaminant in the analyzed samples, because 80% of them exceeded the severe effect level (SEL). The Sungai Linggi river basin is representative of a typical Malaysian river basin. The main river and its tributaries flow through lands that are quite diverse. Upstream is the Seremban township and Senawang industrial areas, whereas, downstream are oil palm plantations and poultry farming operations are more prominent. In addition, this river, being on an estuary, is very significant to aquaculture activities, and is important to aquatic and coastal wildlife. The total mercury concentration found in sediment samples taken from the coastal areas of Malaysia (Straits of Malacca and South China Sea) was between 4.2 and 163 ng/g (Sakamoto et al. 2004).

Levels of mercury are expected to be high in sediment samples collected from the west Peninsular Malaysian coast, because of the hydrocarbon contamination that exists in this area (Zakaria et al. 2000). Such hydrocarbons have S- and O-active sites that bind mercury, and may result in a build-up of mercury levels (CCME 1997). Another source of elemental mercury in sediments and elsewhere is its presence in manometers (commonly used at gas metering sites and at refining and gas plants) (Wilhelm and McArthur 1995).

3.3 *Biological Samples*

The presence of mercury residues in Malaysia has been monitored in a variety of biological organisms and media, including the following: algal and corals species (Sivalingam 1980; Mokhtar et al. 2002), fish and seafood (Babji et al. 1979, 1986; Zahari and Shafie 1987; Law and Singh 1991; Rahman et al. 1997; Yap et al. 2003; Sakamoto et al. 2004; Agusa et al. 2005; Agusa et al. 2007; Kamaruzzaman et al. 2007; Alkarkhi et al. 2008; Hajeb et al. 2009, 2010a, b), food and medical herbs (Wong and Koh 1985; Ang 2004; Ang et al. 2004; Ang and Lee 2005, 2006, 2007; Sharif et al. 2008), and human hair (Sivalingam and Sani 1980; Sarmani et al. 1994; Sarmani and Alakili 2004; Tan et al. 2006; Hajeb et al. 2008; Tengku Hanidza et al. 2008).

3.4 *Algae and Corals*

Few studies exist on mercury residues in algae and corals from Malaysian waters. Coral samples (*Porites* and *Favia* sp.), taken along the shorelines of Tioman and Labuan Island, Teluk Sepangar and Tanjung Aru, were analyzed for mercury residues (Mokhtar et al. 2002). The mercury residues in samples from *Porites* and *Favia* that were collected from marine waters of Tioman Island had levels that ranged from 0.01 to 0.24, and 0.03–0.26 $\mu\text{g/g}$, respectively. Mercury levels in coral samples from other locations were below the detection limit ($<0.01 \mu\text{g/g}$). Mercury contamination levels were investigated in one species of Cyanophyta, fourteen species of the Rhodophyta, five species of the Phaeophyta, and six species of the Chlorophyta from Penang waters (Sivalingam 1980). The total mercury levels detected in algae were $<1.025 \mu\text{g/g}$, indicating that the waters around the island of Penang contained low levels of mercury contamination.

3.5 *Fish and Seafood*

In Table 2, the levels of mercury residues that were identified in several fish species and in seafood samples from Malaysia are presented. The most recent analyses were performed by Hajeb et al. (2009, 2010a, b) and Agusa et al. (2007, 2005), who reported mercury concentrations in marine fish bought from local markets, and which were taken from the west and east coasts of Peninsular Malaysia. Some of these sampled fish species (e.g., tuna, mackerel) contained elevated mercury levels in muscle and liver tissues (Hajeb et al. 2009, 2010a, b). Agusa et al. (2007, 2005) showed that fish samples collected from the west coast retained high mercury levels (0.35–0.37 $\mu\text{g/g}$); similar results (0.778–0.914 $\mu\text{g/g}$) for samples taken from the east coastline were reported in studies performed by Hajeb et al. (2009, 2010a, b). It is therefore apparent that sources of mercury contamination do exist along both coastlines.

Yap et al. (2003) reported mercury levels in the green-lipped mussel (*Perna viridis*) that were collected from the west Peninsular Malaysian coast (viz., 3.89–50.00 $\mu\text{g/g}$ wet wt). Three of the earliest reports on mercury concentration of seafood in Malaysia (Suan and Loong 1981; Babji et al. 1979; Noramly and Marof 1973) also showed that the mercury concentrations that existed in several species of marine fish, prawns, cuttlefish, crab, and molluscs were $<0.5 \mu\text{g/g}$. However, Sivalingam and Sani (1980) reported higher mercury content in several different marine fish species and in cockles (1.34–8.91 $\mu\text{g/g}$) collected in the State of Penang. Rahman et al. (1997) reported that the total mercury levels in seafood samples collected along the west coast (Mersing, Kuala Perils, Batang Tiga, Benut, Kuala Kedah, Kuala Selangor, Sg. Buluh, Morib, and Kuala Juru) were in the range of 0.28–0.61 $\mu\text{g/g}$ (dry wt), whereas samples collected along the east coast (Bachok, Kuala Trengganu, Paka, Marang, Kuantan, Rompin, and Pekan) were in the range

Table 2 Mercury levels found in fish/seafood muscle, liver and heart taken from Malaysian waters

Fish/biota	Location	Tissue	Mercury level ($\mu\text{g/g}$)		Methylmercury level ($\mu\text{g/g}$)		MeHg/THg (%)	Reference
			Dry wt	Wet wt	Dry wt	Wet wt		
Short-bodied mackerel	Market,	Muscle	0.45 \pm 0.06	0.15 \pm 0.03	0.38 \pm 0.11	0.13 \pm 0.09	84.6 \pm 6.5	Hajeb et al. (2010a)
	Selangor	Liver	0.03 \pm 0.02	0.02 \pm 0.01	0.02 \pm 0.01	0.01 \pm 0.02	53.3 \pm 3.5	
Scad	Market,	Muscle	0.04 \pm 0.00	-	-	-	-	Hajeb et al. (2010a)
	Selangor	Liver	0.02 \pm 0.01	0.01 \pm 0.00	-	-	-	
Narrow-barred Spanish mackerel	Market,	Muscle	0.04 \pm 0.02	0.01 \pm 0.01	0.04 \pm 0.03	0.01 \pm 0.01	74.2 \pm 5.3	Hajeb et al. (2010a)
	Selangor	Liver	0.05 \pm 0.03	0.03 \pm 0.02	0.02 \pm 0.01	0.01 \pm 0.01	36.0 \pm 5.1	
Black pomfret	Market,	Muscle	0.13 \pm 0.11	0.04 \pm 0.03	0.06 \pm 0.06	0.02 \pm 0.01	55.3 \pm 9.1	Hajeb et al. (2010a)
	Selangor	Liver	0.10 \pm 0.07	0.06 \pm 0.05	0.03 \pm 0.01	0.02 \pm 0.00	31.6 \pm 3.9	
Greasy grouper	Market,	Muscle	0.03 \pm 0.02	0.01 \pm 0.00	0.02 \pm 0.00	0.01 \pm 0.00	75.0 \pm 2.9	Hajeb et al. (2010a)
	Selangor	Liver	0.03 \pm 0.02	0.01 \pm 0.00	-	-	-	
Long tail tuna	Market,	Muscle	0.50 \pm 0.07	0.12 \pm 0.08	0.42 \pm 0.24	0.10 \pm 0.01	88.5 \pm 6.6	Hajeb et al. (2010a)
	Selangor	Liver	0.49 \pm 0.12	0.12 \pm 0.03	0.25 \pm 0.11	0.06 \pm 0.01	49.1 \pm 2.7	
Chacunda gizzard shad	Market,	Muscle	0.09 \pm 0.02	0.04 \pm 0.02	0.05 \pm 0.03	0.02 \pm 0.01	49.8 \pm 6.8	Hajeb et al. (2010a)
	Selangor	Liver	0.03 \pm 0.01	0.01 \pm 0.00	-	-	-	
Yellow-banded scad	Market,	Muscle	0.06 \pm 0.01	0.03 \pm 0.02	0.04 \pm 0.03	0.02 \pm 0.01	56.1 \pm 5.5	Hajeb et al. (2010a)
	Selangor	Liver	0.04 \pm 0.01	0.03 \pm 0.02	0.01 \pm 0.00	0.01 \pm 0.00	37.7 \pm 6.1	
Eastern little tuna	Market,	Muscle	0.05 \pm 0.04	0.01 \pm 0.01	0.01 \pm 0.00	0.01 \pm 0.00	75.2 \pm 7.3	Hajeb et al. (2010a)
	Selangor	Liver	0.02 \pm 0.01	0.01 \pm 0.00	-	-	-	
Delagoa threadfish	Market,	Muscle	0.09 \pm 0.05	0.03 \pm 0.02	0.06 \pm 0.04	0.02 \pm 0.00	59.3 \pm 4.2	Hajeb et al. (2010a)
	Selangor	Liver	0.09 \pm 0.06	0.03 \pm 0.01	0.03 \pm 0.03	0.01 \pm 0.00	40.1 \pm 3.5	
Giant Perch	Market,	Muscle	0.10 \pm 0.06	0.02 \pm 0.02	0.09 \pm 0.06	0.02 \pm 0.00	81.2 \pm 8.6	Hajeb et al. (2010a)
	Selangor	Liver	0.01 \pm 0.00	0.01 \pm 0.00	-	-	-	
Sardine	Market,	Muscle	-	-	-	-	-	Hajeb et al. (2010a)
	Selangor	Liver	0.01 \pm 0.01	0.01 \pm 0.00	-	-	-	
Short-bodied mackerel	Kuantan,	Muscle	-	0.279 \pm 0.021	-	0.219 \pm 0.014	70 \pm 5.2	Hajeb et al. (2010a)
	Pahang	Liver	-	0.897 \pm 0.087	-	0.388 \pm 0.043	40 \pm 3.6	
Short-bodied mackerel	Chendring,	Muscle	-	0.778 \pm 0.074	-	0.665 \pm 0.054	82 \pm 8.2	Hajeb et al. (2010a)
	Terengganu	Liver	-	1.357 \pm 0.481	-	0.616 \pm 0.062	46 \pm 4.3	

Short-bodied mackerel	Kuala Perlis, Kedah	Muscle Liver	- -	0.229±0.011 0.582±0.064	- -	0.179±0.018 0.242±0.024	76±7.1 41±3.2	Hajeb et al. (2010a)
Long-tail tuna	Kuantan, Pahang	Muscle Liver	- -	0.446±0.051 1.463±0.189	- -	0.405±0.030 0.619±0.081	78±8.0 43±2.2	Hajeb et al. (2010a)
Long-tail tuna	Chendring, Terengganu	Muscle Liver	- -	0.914±0.066 1.386±0.261	- -	0.708±0.056 0.651±0.058	76±6.7 47±4.4	Hajeb et al. (2010b)
Long-tail tuna	Kuala Perlis, Kedah	Muscle Liver	- -	0.225±0.045 0.583±0.044	- -	0.187±0.009 0.236±0.018	81±9.6 45±5.7	Hajeb et al. (2010a)
Kawakawa	Cabang Tiga, Kelantan	Muscle Liver	0.09 0.10	- -	- -	- -	- -	Agusa et al. (2007)
Torpedo scad	Kuala Terengganu	Muscle Liver	0.37 0.13	- -	- -	- -	- -	Agusa et al. (2007)
Bigeye scad	Langkawi	Muscle Liver	0.13 0.12	- -	- -	- -	- -	Agusa et al. (2007)
Redtail scad	Langkawi	Muscle Liver	0.11 0.10	- -	- -	- -	- -	Agusa et al. (2007)
Indian mackerel	Mersing, Johor	Muscle Liver	<0.05 0.12	- -	- -	- -	- -	Agusa et al. (2007)
Javelin grunter	Mersing, Johor	Muscle Liver	0.09 0.06	- -	- -	- -	- -	Agusa et al. (2007)
Doublespotted queenfish	Parit Jawa, Johor	Muscle Liver	0.09 0.36	- -	- -	- -	- -	Agusa et al. (2007)
Black pomfret	Port Dickson, Johor	Muscle Liver	0.05 0.21	- -	- -	- -	- -	Agusa et al. (2007)
Doublelined tonguesole	Langkawi	Muscle Liver	0.09 0.24	- -	- -	- -	- -	Agusa et al. (2007)
Shortfin scad	Langkawi	Muscle Liver	0.06 <0.05	- -	- -	- -	- -	Agusa et al. (2007)
Yellowfin seabream	Langkawi	Muscle Liver	0.31 0.35	- -	- -	- -	- -	Agusa et al. (2007)

(continued)

Table 2 (continued)

Fish/biota	Location	Tissue	Mercury level ($\mu\text{g/g}$)		Methylmercury level ($\mu\text{g/g}$)		MeHg/THg (%)	Reference
			Dry wt	Wet wt	Dry wt	Wet wt		
Two-spotted Glass catfish	Bebar river	Muscle	0.05–0.32	–	–	–	–	Kamaruzzaman et al. (2007)
Long-whisker catfish	Bebar river	Muscle	0.12–0.43	–	–	–	–	Kamaruzzaman et al. (2007)
Banded leaf fish	Bebar river	Muscle	0.01–0.14	–	–	–	–	Kamaruzzaman et al. (2007)
Hard-lipped barb	Bebar river	Muscle	0.05–0.09	–	–	–	–	Kamaruzzaman et al. (2007)
<i>Scolopsis monogramma</i>	Port Dickson	Muscle	–	0.168	–	0.147	87.5	Sakamoto et al. (2004)
<i>Lutjanus johnii</i>	Port Dickson	Muscle	–	0.053	–	0.042	79.2	Sakamoto et al. (2004)
Mempinang	Port Dickson	Muscle	–	0.043	–	0.034	79.1	Sakamoto et al. (2004)
Mempinang	Port Dickson	Muscle	–	0.043	–	0.034	79.1	Sakamoto et al. (2004)
<i>Siganus canaliculatus</i>	Port Dickson	Muscle	–	0.004	–	0.004	100.0	Sakamoto et al. (2004)
<i>Scatohagus argus</i>	Port Dickson	Muscle	–	0.042	–	0.034	81.0	Sakamoto et al. (2004)
Pfifax	Port Dickson	Muscle	–	0.017	–	0.015	88.2	Sakamoto et al. (2004)
<i>Siganus jayus</i>	Port Dickson	Muscle	–	0.010	–	0.009	90.0	Sakamoto et al. (2004)
Crab	Port Dickson	Muscle	–	0.163	–	0.146	89.6	Sakamoto et al. (2004)
Gnathonodom species	Terengganu	Muscle	–	0.056	–	0.046	82.1	Sakamoto et al. (2004)
<i>Chiloscyllium</i> sp.	Terengganu	Muscle	–	0.229	–	0.154	67.2	Sakamoto et al. (2004)
<i>Sardinella fimbriata</i>	Terengganu	Muscle	–	0.042	–	0.034	81.0	Sakamoto et al. (2004)
<i>Rastrelliger kanagurta</i>	Terengganu	Muscle	–	0.018	–	0.014	77.8	Sakamoto et al. (2004)
Sciaenidae	Terengganu	Muscle	–	0.043	–	0.026	60.5	Sakamoto et al. (2004)
<i>Lutjanus malabaricus</i>	Terengganu	Muscle	–	0.021	–	0.016	76.2	Sakamoto et al. (2004)
<i>Nemipterus furcosus</i>	Terengganu	Muscle	–	0.191	–	0.157	82.2	Sakamoto et al. (2004)
<i>Laligo</i> sp.	Terengganu	Muscle	–	0.019	–	0.021	110.5	Sakamoto et al. (2004)
<i>Euthynnus affinis</i>	Terengganu	Muscle	–	0.024	–	0.017	70.8	Sakamoto et al. (2004)
<i>Penaeidae</i>	Terengganu	Muscle	–	0.010	–	0.009	90.0	Sakamoto et al. (2004)
<i>Portunus pelagicus</i>	Terengganu	Muscle	–	0.013	–	0.008	61.5	Sakamoto et al. (2004)
Green-lipped mussel (<i>Perna viridis</i>)	West coast	–	0.003–0.050	–	–	–	–	Yap et al. (2003)
Indian mackerel	Mersing	Muscle	0.45 ± 0.01	–	0.30 ± 0.0	–	67	Rahman et al. (1997)

Spanish mackerel	Bachok	Muscle	0.21 ± 0.01	-	0.17 ± 0.02	-	81	Rahman et al. (1997)
	Kuala Trengganu	Muscle	0.21 ± 0.01	-	0.19 ± 0.05	-	90	Rahman et al. (1997)
	Paka	Muscle	0.33 ± 0.03	-	0.22 ± 0.05	-	67	Rahman et al. (1997)
	Marang	Muscle	0.23 ± 0.02	-	0.17 ± 0.02	-	74	Rahman et al. (1997)
	Mersing	Muscle	0.43 ± 0.05	-	0.36 ± 0.01	-	85	Rahman et al. (1997)
	Kuala Perlis	Muscle	0.33 ± 0.03	-	0.31 ± 0.04	-	94	Rahman et al. (1997)
	Batang Tiga	Muscle	0.32 ± 0.02	-	0.20 ± 0.02	-	63	Rahman et al. (1997)
	Penut	Muscle	0.30 ± 0.02	-	0.19 ± 0.03	-	63	Rahman et al. (1997)
	Kuala Kedah	Muscle	0.61 ± 0.04	-	0.49 ± 0.04	-	80	Rahman et al. (1997)
	Kuala Perlis	Muscle	0.51 ± 0.02	-	0.29 ± 0.05	-	57	Rahman et al. (1997)
Squid (<i>Loligo</i> sp.)	Mersing		0.27 ± 0.03	-	0.16 ± 0.03	-	59	Rahman et al. (1997)
	Kuantan		0.43 ± 0.08	-	0.30 ± 0.04	-	70	Rahman et al. (1997)
Shrimp (<i>Penaeus</i> sp.)	Pekan		0.21 ± 0.01	-	0.12 ± 0.05	-	57	Rahman et al. (1997)
	Kuala Selangor		0.29 ± 0.05	-	0.23 ± 0.05	-	79	Rahman et al. (1997)
	Kuantan	Muscle	0.37 ± 0.05	-	0.21 ± 0.03	-	57	Rahman et al. (1997)
	Kuala Kedah	Muscle	0.38 ± 0.01	-	0.17 ± 0.02	-	45	Rahman et al. (1997)
Cockle (<i>Anadara granosa</i>)	Rompin	Muscle	0.21 ± 0.01	-	0.16 ± 0.03	-	76	Rahman et al. (1997)
	Mersing	Muscle	0.28 ± 0.05	-	0.15 ± 0.03	-	54	Rahman et al. (1997)
	Sg. Buluh	Muscle	0.57 ± 0.03	-	0.46 ± 0.04	-	81	Rahman et al. (1997)
	Morib	Muscle	0.28 ± 0.04	-	0.20 ± 0.02	-	71	Rahman et al. (1997)
	Kuala Juru	Muscle	0.55 ± 0.02	-	0.44 ± 0.06	-	80	Rahman et al. (1997)
	Parrot wrasse	Muscle	2.46	-	-	-	-	Sivalingam and Sani (1980)
Sardine	Liver		7.07	-	-	-	-	Sivalingam and Sani (1980)
	Heart		8.73	-	-	-	-	Sivalingam and Sani (1980)
	Muscle		1.91	-	-	-	-	Sivalingam and Sani (1980)
	Liver		7.12	-	-	-	-	Sivalingam and Sani (1980)
	Heart		8.91	-	-	-	-	Sivalingam and Sani (1980)

(continued)

Table 2 (continued)

Fish/biota	Location	Tissue	Mercury level ($\mu\text{g/g}$)		Methylmercury level ($\mu\text{g/g}$)		Reference
			Dry wt	Wet wt	Dry wt	Wet wt	
Sole	Kuala Juru	Muscle	1.90	-	-	-	Sivalingam and Sani (1980)
		Liver	7.74	-	-	-	
Mullet	Kuala Juru	Heart	4.47	-	-	-	Sivalingam and Sani (1980)
		Muscle	1.34	-	-	-	
		Liver	4.02	-	-	-	
		Heart	3.80	-	-	-	
Catfish	Kuala Juru	Muscle	1.88	-	-	-	Sivalingam and Sani (1980)
		Liver	4.03	-	-	-	
		Heart	5.56	-	-	-	
Silver pomfret	Sungei Kelang	Muscle	-	0.05	-	-	Suan and Loong (1981)
Grass toadfish	Sungei Kelang	Muscle	-	0.01	-	-	Suan and Loong (1981)
catfish	Sungei Kelang	Muscle	-	0.11	-	-	Suan and Loong (1981)
		Muscle	-	0.03	-	-	Suan and Loong (1981)
Anchovy	Sungei Kelang	Muscle	-	0.02	-	-	Suan and Loong (1981)
		Muscle	-	0.02	-	-	Suan and Loong (1981)
Spotted scat	Sungei Kelang	Muscle	-	0.24	-	-	Suan and Loong (1981)
		Muscle	-	0.04	-	-	Suan and Loong (1981)
Sardine	Sungei Kelang	Muscle	-	0.01	-	-	Suan and Loong (1981)
		Muscle	-	0.09	-	-	Suan and Loong (1981)
Striped eel catfish	Perlis	Muscle	-	0.07	-	-	Babji et al. (1979)
		Muscle	-	0.08	-	-	Babji et al. (1979)
Grouper	Kedah	Muscle	-	0.08	-	-	Babji et al. (1979)
		Muscle	-	0.08	-	-	Babji et al. (1979)
Eel catfish	Penang	Muscle	-	0.09	-	-	Babji et al. (1979)
		Muscle	-	0.10	-	-	Babji et al. (1979)
Croaker	Selangor	Muscle	-	0.09	-	-	Babji et al. (1979)
		Muscle	-	0.10	-	-	Babji et al. (1979)
Sillagos	Terengganu	Muscle	-	0.10	-	-	Babji et al. (1979)
		Muscle	-	0.10	-	-	Babji et al. (1979)
Goatfish	Perlis	Muscle	-	0.10	-	-	Babji et al. (1979)
		Muscle	-	0.10	-	-	Babji et al. (1979)
Dart	Perlis	Muscle	-	0.10	-	-	Babji et al. (1979)

MeHg methylmercury, *THg* total mercury

of 0.21–0.43 $\mu\text{g/g}$. Most study results have revealed that higher mercury residues exist in seafood taken from the west coast than from the east coast of Peninsular Malaysia. The fact that the west coast is more industrialized probably accounts for this difference, as does the fact that industrial sources and agricultural activities along the west coast are near fishing areas, wherein mercury may exist in marine sediment and be available for transfer to fish (Rahman et al. 1997).

Only a few studies have been performed on the methylmercury content in fish and seafood in Malaysia (Hajeb et al. 2010a, b; Rahman et al. 1997). These authors found that in a variety of samples studied, methylmercury residues were in the range of 45–94% of total mercury residues. Tuna and mackerel were reported to have a higher level of the organic mercury as compared to other species. The highest ratio of organic mercury (94%) was found in Spanish mackerel collected from Kuala Perlis (Rahman et al. 1997), short bodied mackerel (89%) and long tail tuna (91.5%) collected from Terengganu and Pahang. This may suggest the presence of some point sources of mercury contamination in Peninsular Malaysia, or in relevant open water areas where the fish were caught.

4 Mercury Contamination of Food, Herbs, and Medicines

Heavy metal poisoning such as with mercury has long been associated with traditional medicines (Ang and Lee 2007). Malaysia has established a maximum level of $<0.5 \mu\text{g/g}$ for the presence of heavy metals in traditional medicinal preparations, with particular reference to the presence of mercury (Jaafar 1995). Because mercury is recognized as a reproductive toxicant, its concentration in some traditional medicines and herbal products has been evaluated in Malaysia. Wong and Koh (1985) studied the amounts of mercury in 99 common Chinese medicines that are available in Malaysian markets. These authors showed that about 7% of analyzed samples contained mercury residues of more than 1,000 $\mu\text{g/g}$, 6% had 0.5–20 $\mu\text{g/g}$, and 85% had less than 0.5 $\mu\text{g/g}$ of mercury. However, they stated that the source of mercury was probably HgS, which is less toxic than other mercury compounds; HgS is also highly insoluble and is thus more likely to be excreted after ingestion.

In a survey of the mercury content of herbal preparations that contained Tongkat Ali hitam (*Eurycoma longifolia*) in the Malaysian market, 15% of the tested products contained 0.62–2.32 $\mu\text{g/g}$ of mercury. The results of this study showed that 85% of the products tested complied with the Malaysian quality requirement ($<0.5 \mu\text{g/g}$) for the presence of mercury in traditional medicines; however, such preparations could not be assumed to be safe from mercury contamination because of batch-to-batch inconsistency (Ang et al. 2004).

The mercury content of 100 pharmaceutical dosage forms of *Smilax luzonensis* (greenbriers), which is eaten as an aphrodisiac in the Malaysian community, was tested. The results showed that 86% of the products complied with the quality requirement for traditional medicinal preparations in Malaysia, in which mercury content was of particular concern. However, mercury was detected in 14% of the

products, which resulted in a call for urgent action by the Malaysian government to rectify the abnormal amounts of contamination found (Ang and Lee 2005).

Ang and Lee (2006) determined the level of mercury in 100 products from Malaysia of a black variety of Tongkat Ali that had been prepared into various pharmaceutical dosage forms of the herbal preparation. The results showed that 26% of the products contained 0.53–2.35 $\mu\text{g/g}$ of mercury. Therefore, the authors concluded that those products do not comply with the quality requirement for traditional medicines in Malaysia (i.e., $<0.5 \mu\text{g/g}$) (Jaafar 1995). In another survey of mercury content of 100 herbal products containing *Smilax myosotiflora* from the Malaysian market, 11% of the examined products exceeded 0.5 $\mu\text{g/g}$ of mercury (Ang and Lee 2007). The presence of mercury in traditional medicines may derive from growing the medicinal plants from which the medicines come in seriously polluted soil; alternatively, these products may contain animal and/or mineral material that are contaminated with heavy metals (Chuang et al. 2000). Samples of locally processed raw Malaysian food products (e.g., salted fish, shrimp paste, and dried shrimps) that are widely used as main ingredients in local cooking were collected from Malacca (main production and distribution center for these foods) and were then analyzed for mercury contamination. Results indicated that these samples did not contain detectable mercury (Sharif et al. 2008).

5 Human Health Indicators

Few studies have addressed that mercury levels may exist in human tissues and fluids (hair and urine) in the Malaysian population. Having such information would help assess the human exposure levels to mercury. The mercury levels that exists in urine taken from dentists and dental auxiliaries, and the relationship that the Hg levels have to the number of amalgam fillings, type of amalgam used, the work load, and mercury exposures from seafood and other activities were studied by Tan et al. (2006). Information about the potential degree of exposures to mercury (i.e., from work, seafood consumption, and other exposures) was obtained from a questionnaire survey. The urinary mercury level found by Tan et al. (2006) was $3.19 \pm 6.61 \mu\text{g/L}$, with no significant differences among different staff categories. Less than 0.5% of respondents had higher urinary mercury levels than the guidance value of 20 $\mu\text{g/L}$, and only 0.21% of respondents had urine that exceeded 50 $\mu\text{g/L}$. The levels in urine of oral healthcare personnel did not show any significant association with the frequency of intake of seafood, or with amalgam status (number of amalgam fillings), type of alloy used, or amalgam workload. However, Hg urine levels were associated with increased reports of personnel medical symptoms.

In an earlier study (Sivalingam and Sani 1980), the total mercury level in hair samples from residents of a few fishing communities in the state of Penang was reported as being between 7.36 and 16.10 $\mu\text{g/g}$. There was no correlation between mercury levels in hair of the studied populations and mercury concentrations in local fish samples. Later, Sarmani et al. (1994) analyzed for mercury in hair samples

collected from fishermen and their families residing in an industrialized area in Penang, and in a nonindustrialized area in Terengganu. The mercury levels in the hair samples of residents from Penang and ranged from 0.45 to 16.68 $\mu\text{g/g}$, and those of Terengganu from 6.79 to 18.31 $\mu\text{g/g}$. These study results demonstrated that mercury levels in human hair do depend on the pattern of fish consumption. Sarmani and Alakili (2004) found that the contamination levels in hair samples of Malaysian citizens of Kuala Lumpur were 3.38 and 1.13 $\mu\text{g/g}$ for total mercury, and methylmercury, respectively. The authors of this study identified fish consumption as a significant route of mercury exposure.

Hajeb et al. (2008), in a more extensive study, surveyed hair mercury levels in the rural and urban communities of four coastal states of Malaysia: Kedah, Terengganu, Johor, and Selangor. The mercury levels found ranged from 0.01 to 21.00 $\mu\text{g/g}$ dry wt. The authors reported a significant positive correlation between hair mercury concentration and fish consumption in the residents of all four communities studied. The mercury exposure to residents of rural communities was higher than those in urban areas. However, the mercury levels found in residents of all the studied communities was much lower than the WHO's no observable adverse effect level (NOAEL) (50 $\mu\text{g/g}$ dry wt). Tengku Hanidza et al. (2008) studied the concentration of mercury in hair samples collected from residents of two rural, and two urban coastal communities, Yan and Alor Setar in the state of Kedah, and Bachok and Kota Bharu in the state of Kelantan, respectively. The geometric means for total mercury levels found in those communities were as follows: 1.38 $\mu\text{g/g}$ dry wt (Yan), 1.20 $\mu\text{g/g}$ dry wt (Alor Setar), 1.24 $\mu\text{g/g}$ dry wt (Bachok), and 1.07 $\mu\text{g/g}$ dry wt (Kota Bharu). Two persons, each from Alor Setar and Kota Bharu, had a high total mercury level in hair (223.58 and 803.16 $\mu\text{g/g}$ dry wt, respectively). Their analysis for methyl mercury showed that the levels were within 1.36 and 1.91 $\mu\text{g/g}$ dry wt, respectively. Age and fish consumption appeared to have a significant effect on levels of hair mercury levels in those populations.

6 Summary

Although several studies have been published on levels of mercury contamination of the environment, and of food and human tissues in Peninsular Malaysia, there is a serious dearth of research that has been performed in East Malaysia (Sabah and Sarawak). Industry is rapidly developing in East Malaysia, and, hence, there is a need for establishing baseline levels of mercury contamination in environmental media in that part of the country by performing monitoring studies. Residues of total mercury and inorganic mercury in food samples have been determined in nearly all previous studies that have been conducted; however, few researchers have analyzed samples for the presence of methylmercury residues. Because methylmercury is the most toxic form of mercury, and because there is a growing public awareness of the risk posed by methylmercury exposure that is associated with fish and seafood consumption, further monitoring studies on methylmercury in food are also

essential. From the results of previous studies, it is obvious that the economic development in Malaysia, in recent years, has affected the aquatic environment of the country. Primary areas of environmental concern are centered on the rivers of the west Peninsular Malaysian coast, and the coastal waters of the Straits of Malacca, wherein industrial activities are rapidly expanding. The sources of existing mercury input to both of these areas of Malaysia should be studied and identified.

Considering the high levels of mercury that now exists in human tissues, efforts should be continued, and accelerated in the future, if possible, to monitor mercury contamination levels in the coastal states, and particularly along the west Peninsular Malaysian coast. Most studies that have been carried out on mercury residues in environmental samples are dated, having been conducted 20–30 years ago; therefore, the need to collect much more and more current data is urgent. Furthermore, establishing baseline levels of mercury exposure to humans in Malaysia will be useful in establishing the levels at which detrimental effects in both humans and marine life may occur, and therefore the levels at which warnings should be raised or limits established. In particular, we believe that two or three monitoring centers should be established in Peninsular Malaysia, and one in East Malaysia for the specific purpose of monitoring for the presence of hazardous environmental chemicals, and particularly monitoring for heavy metals such as mercury that reach food that is subject to consistent human consumption.

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Genotoxic and Reprotoxic Effects of Tritium and External Gamma Irradiation on Aquatic Animals

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1 Introduction

Aquatic systems are inhabited by a large variety of species, several of which comprise important components in human diets. Aquatic systems are also the final receptors of a whole range of pollutants, including radioactive ones, because the majority of nuclear facilities are connected to either rivers or to the marine environment.

The main radionuclides routinely released from nuclear power plants and nuclear fuel reprocessing plants are gamma (γ) emitters (e.g., ^{137}Cs , ^{60}Co , ^{54}Mn) and tritium. The latter is the most abundantly released radionuclide from the nuclear industry (around 10^{16} Bq/year; Adam-Guillermin et al. 2010). In the near future, the release of tritium is expected to increase with the implementation of new reactors (e.g., European Pressurized Reactor or EPR) and the development of the ITER (International Thermonuclear Experimental Reactor) nuclear fusion facility, which will enhance public concerns about this radionuclide. Tritium is a radioactive isotope of hydrogen. It behaves chemically like hydrogen, forming water molecules, dihydrogen gas, or biomolecules. It is a very low-energy beta emitter (average energy of 5.7 keV) of short range (average track length of 0.56 μm in water). As a result, the average ionization density (and linear energy transfer) produced by the emitted beta particle is significantly higher than that produced by higher energy particles or photons, such as ^{60}Co (HPA 2007). In addition, in situ transmutation of tritium into helium and enrichment of water in the DNA hydration shell contribute to the enhancement of tritium effects on DNA (HPA 2007).

Radionuclide exposure may cause major alterations to the structure and function of biological macromolecules, such as lipids, carbohydrates, proteins, and nucleic acids. Assessing DNA damage is important because such damage may produce irreversible effects such as carcinogenesis (Stein et al. 1994; Wirgin et al. 1994) and teratogenesis (Theodorakis et al. 1997). It can also affect fecundity (Anderson and Wild 1994; Theodorakis et al. 1997), immune function (Hurks et al. 1995), or deplete cellular energy stores (Pieper et al. 1999). Another important aspect of DNA damage is that it can also contribute to evolutionary effects by transmitting mutations to subsequent generations (Frankham 2005), partially through epigenetic mechanisms (Aypar et al. 2011). Consequently, effects on DNA, measured at the subcellular level, should theoretically be correlated to effects on individuals, populations, or communities.

Although some literature reviews have focused on the genotoxicity of pollutants in aquatic organisms (Mitchelmore and Chipman 1998; Jha 2004), none have specifically addressed the genotoxicity of radionuclides, and the associated effects they have at higher organizational levels. Importantly, our review provides an additional analysis of the data by fitting nonlinear curves (described below) to the dose–response data provided in the reviewed manuscripts. The additional analyses allowed us to calculate several endpoints that are commonly associated with ecotoxicological studies, such as the 10% effect dose rate (EDR_{10}), and to use such endpoints as a common metric for comparing the data published in the literature. Such a review is important because of the routine releases into aquatic environments

that occur from nuclear power plants, the large releases into the marine environment from the Fukushima accident (Garnier-Laplace et al. 2011), and because of the renewed debate on the pros and cons of nuclear energy production (e.g., Ferguson 2011). In this review, we examine the current state-of-knowledge concerning the effects of radionuclides on DNA integrity, reproductive ability, teratogenesis, and the early life-stage survival of aquatic organisms.

2 Methods Used

We limited the scope of this review to studies of animals whose primary or critical life stage is aquatic, and to animals which were exposed to γ irradiation or tritium in the laboratory or in the field. We tried to take into account the papers published to date in French or in English, including the “grey literature,” such as reports that utilize data extracted from the Frederica database (Coppstone et al. 2008). Special attention was given to dose assessment, as it is generally the weakest point of such studies. Dose, as used in radiological studies, does not have the same units as that used for nonradioactive contaminants (e.g., mg stable Hg/kg fish mass). Instead, dose (in Gray, Gy) from a radioactive contaminant perspective refers to the energy (Joules) absorbed per mass (kg) of tissue when a radionuclide undergoes radioactive decay (i.e., 1 J/kg = 1 Gy). When dealing with dose rate, the international system of unit is Gy per unit time. The background dose rate from natural radioactivity is a few $\mu\text{Gy/day}$, but can be enhanced by naturally occurring radionuclides in soil or by cosmic radiation as the elevation above the earth’s surface increases (Gómez-Ros et al. 2004). Dose rates may be increased due to the nuclear fuel cycle. For example, the maximal absorbed dose rate in aquatic organisms (macrophytes) from the Cumbrian coast in the UK was estimated to be 96 $\mu\text{Gy/day}$ (Coppstone et al. 2001).

In the reviewed papers, radiological doses were sometimes calculated by the original authors; if not, we estimated dose rate according to the following equation (modified from Hagger et al. 2005):

$$D_{\beta} = 5.76 \times 10^{-7} \times \epsilon_{\beta} \times C, \quad (1)$$

where D_{β} is the dose rate in Gy/h; 5.76×10^{-7} is a conversion factor; ϵ_{β} is the average beta-ray energy ($^3\text{H} = 0.00569 \text{ MeV}$), and C is the concentration of tritium (Bq/mL). These calculations were based on the assumptions that: (a) ^3H was uniformly distributed within the organism over the exposure period and (b) no concentration of ^3H above a water equilibrium level occurred (concentration factor of 1).

The biological endpoints we examined were genotoxicity, reproduction, and development. When dose–response relationships were studied, we attempted to synthesize the results by using standardized critical indices of ecotoxicity such as:

- HNEDR (highest no effect dose rate): highest dose rate for which no statistically significant effect was observed as compared to the control group.

- LOEDR (lowest observed effect dose rate): lowest dose rate for which a statistically significant effect was observed as compared to the biological response in the control group.
- EDR_x, or effective dose rate $x\%$ corresponding to the dose rate giving an $x\%$ effect as compared to the control group. In the same manner, LDR_x or lethal dose rate $x\%$ corresponds to the dose rate to give an $x\%$ mortality as compared to the control group. EDR_x values were estimated by fitting a log-logistic nonlinear regression to quality-assessed data sets from the Frederica database and from the available literature. To be acceptable for modeling, datasets had, at first, to satisfy several criteria (described in Garnier-Laplace et al. 2010). Then, the estimated EDR_x had to be bracketed by experimental points. Nonlinear regressions were calculated using a log-logistic model (Ritz and Streibig 2005) with the R software (R Development Core Team 2009) and the “drc” add-on package. Because EDR₁₀ values were obtained using more robust methods, it was the preferred endpoint to LOEDR, when both values were available.

Some reported research was specifically performed to compare the different efficiencies of tritium and γ rays, and the resulting values were described by using the relative biological effectiveness (RBE) index. RBE quantifies the different efficiencies of radiation types to produce a biological effect. It is defined as the ratio of the absorbed dose of the reference radiation to the absorbed dose of the test radiation that is required under similar conditions to produce an identical level of biological response in a particular animal or cellular study. Thus, in human radiobiology, RBEs are used as quality factors to normalize the dose among different forms of radiation that possess different efficiencies. In this chapter, some RBEs could be calculated using ratios of either LOEDR or EDR₁₀ for HTO (tritiated water) and γ rays. Some values were also calculated by the original authors.

3 Review of Tritium Effects

3.1 *Aquatic Invertebrates*

Most research performed on tritium effects to aquatic organisms used HTO (one study was also performed with organic tritium) and studied effects in marine species (Pacific oyster, brine shrimp, blue mussel, polychaete worms, goose barnacle). Only one study has been performed on freshwater organisms, which used daphnids as a biological model (Table 1).

3.1.1 Effects on DNA

As could be expected from the characteristics of beta particles emitted by tritium, DNA alterations were observed in organisms (blue mussels) exposed to tritiated

Table 1 Effects induced in aquatic invertebrates by chronic tritium chronic. Effect dose rate in mGy/day ($\mu\text{Gy/h}$)

Reference	Species	Dose rates	Exposure duration	Studied effect	Endpoint	EDR ₁₀ (mGy/day) ($\mu\text{Gy/h}$)	EDR ₅₀ (mGy/day) ($\mu\text{Gy/h}$)	LOEDR (mGy/day) ($\mu\text{Gy/h}$)	HNEDR (mGy/day) ($\mu\text{Gy/h}$)	RBE
Hagger et al. (2005)	<i>Mytilus edulis</i> (blue mussel, embryo-larvae)	0, 0.03, 0.29, 2.9, 29 mGy/day	24 h	Genotoxicity	Chromosome aberrations	2×10^{-4} (0.009)		0.3 (12) 0.03 (1.2) 0.03 (1.2)		
				Development	Abnormalities in larvae after 72 h	0.03 (1.3) 0.017 (0.7)	0.75 (3.1) 0.032 (1.3)			
				Mortality	Embryo mortality after 72 h	10^{-4} (0.004)	0.010 (0.4)			
					48 h embryo mortality		0.01 (0.4) ^a			
Gudkov and Kipnis (1996)	<i>Daphnia magna</i> (daphnid)	0, 39 nGy/day, 39 $\mu\text{Gy/day}$, 39 mGy/day ^b	71 days	Life history traits over five generations	Fecundity parameters	0.007 (0.3)	0.04 (1.6)			
				Development	Development abnormalities	0.08 (3)	0.04 (1.6)			
Abbott and Mix (1979)	<i>Pollicipes polymerus</i> (goose barnacle, larvae)	0, 190 nGy/day, 1.5, 15, 150 $\mu\text{Gy/day}$, 1.5 mGy/day ^b	32 days	Development	Molting index of post-stage I larvae	8×10^{-4} (0.03)		0.015 (0.62)		
Jaeschke et al. (2011)	<i>Mytilus edulis</i> (blue mussel, adult hemocytes)	0, 0.12 (tritiated glycine), 1.9 and 2.9 mGy/day HTO	7 and 14 days	Genotoxicity	Micronuclei and Comet assay			0.12 (5)		
Jha et al. (2005)	<i>Mytilus edulis</i> (blue mussel adult)	0, 0.3, 3, 12 mGy/day	96 h	Genotoxicity in hemocytes	Micronuclei and Comet assay	0.53 (22)		0.3 (12)		
Nelson (1971)	<i>Crassostrea gigas</i> (Pacific oyster, larvae)	0, 0.0003, 0.003, 0.03, 3, 30 mGy/day	48 h	Development	Development abnormalities	2.8 (117)		3 (125)		
				Mortality	Mortality				30 (1,250)	

(continued)

Table 1 (continued)

Reference	Species	Dose rates	Exposure duration	Studied effect	Endpoint	EDR ₁₀ (mGy/day) (μ Gy/h)	EDR ₃₀ (mGy/day) (μ Gy/h)	LOEDR (mGy/day) (μ Gy/h)	HNEDR (mGy/day) (μ Gy/h)	RBE
Knowles and Greenwood (1997)	<i>Ophryotrocha diadema</i> (polychaete worm, eggs to adults)	0, 175 mGy/day	11 weeks	Reproduction Mortality	Reproduction Mortality of eggs			175 (7,292) idem		1 ^c
Higuchi et al. (1980)	<i>Artemia salina</i> (brine shrimp, eggs to adults)	0, 291 mGy/day, 1.46, 2.91, 10.2 and 21.8 Gy/day	25 days	Reproduction Mortality	Reproduction Hatchability of encysted dry eggs			290 (12,083)	22 × 10 ³ (9.1 × 10 ⁵) 10 ⁴ (4.6 × 10 ⁵)	

Gy/h, dose (in Gray, Gy) from a radioactive contaminant perspective refers to the energy (Joules) absorbed per mass (kg) of tissue when a radionuclide undergoes radioactive decay (i.e., 1 J/kg = 1 Gy). Gy per hour corresponds to the dose rate, HNEDR (highest no effect dose rate): highest dose rate for which no statistically significant effect was observed as compared to the control group, LOEDR (lowest observed effect dose rate): lowest dose rate for which a statistically significant effect was observed as compared to the biological response in the control group, EDRx, or effective dose rate $x\%$ corresponding to the dose rate giving an $x\%$ effect as compared to the control group. In the same manner, LDRx or lethal dose rate $x\%$ corresponds to the dose rate to give an $x\%$ mortality as compared to the control group, RBE (relative biological effectiveness): with this index, one quantifies the different efficiencies of radiation types that produce a biological effect. It is defined as the ratio of the absorbed dose of the reference radiation to the absorbed dose of the test radiation that is required under similar conditions to produce an identical level of biological response in a particular animal or cellular study. Thus, in human radiobiology, RBEs are used as quality factors to normalize the dose among different forms of radiation that possess different efficiencies. In this chapter, RBEs were calculated using ratios of either LOEDR or EDR₁₀ for HTO (tritiated water) and γ rays. Some values were also calculated by the original authors

^aValue given by the authors

^bDose rate calculated from (1)

^cValue given by the authors

water or to organic tritium, either at the egg stage (Hagger et al. 2005) or as adults (Jha et al. 2005; Jaeschke et al. 2011). DNA alterations, determined by using RAPD (randomly amplified polymorphic DNA) profiles and the comet assay, occurred at a low dose rate of 0.3 mGy/day, and progressively increased in a dose-dependent manner at higher rates, notwithstanding the studied life-stage. From the same low dose rate (i.e., 0.3 mGy/day), cytogenetic alterations were also observed, in terms of sister chromatid exchanges (SCEs) and chromosomal aberrations (Hagger et al. 2005). The increase in chromosomal aberrations was not a function of dose, probably because apoptosis mechanisms dominated at the highest doses. The main aberrations observed were acentric fragments that resulted from chromosome or chromatid breaks. The increase in SCE, dependent on the S phase of the cell cycle, suggests that tritium may interfere with the replication processes. DNA alterations were correlated with a significant decrease of normal embryo-larvae (and correspondingly, an increase in mortality; Hagger et al. 2005). Similar results were obtained for adult mussels, in which a significant genotoxicity occurred at even the lowest dose tested (0.3 mGy/day), using micronuclei (MN) frequency and the comet assay (Jha et al. 2005). Interestingly, this genotoxicity was enhanced for organic tritium (Jaeschke et al. 2011). Hence, tritiated glycine was found to be 15 times more genotoxic than tritiated water in adult blue mussels, exposed at dose rates from 0.12 mGy/day (glycine form) to 3 mGy/day (tritiated water).

3.1.2 Effects on Survival of Early Life Stages and Reproduction

Invertebrate survival was affected at low dose rates, but only following long exposure periods. Hence, chronic exposure of daphnids up to a dose rate of 39 mGy/day did not induce any mortality following 72 h of exposure (Gudkov and Kipnis 1996), whereas exposure for five generations (71 days) led to mortality in all generations for dose rates of 39 μ Gy/day and 39 mGy/day. A dose rate-dependent reduction of life span was observed, with a decrease of 50% at the highest dose rate (39 mGy/day) for the first generation.

The sensitivity of marine organisms to tritium varies by species and endpoints considered. Survival of 48-h-old Pacific oyster larvae was not altered following their exposure during the egg stage to low dose rates of HTO (from 0.003 to 30 mGy/day) (Nelson 1971). Brine shrimp, known as a radioresistant animal, was exposed to HTO at dose rates from 291 mGy/day to 21.8 Gy/day to study their growth, survival, and fecundity (Higuchi et al. 1980). Their life span was significantly reduced (30%) at dose rates greater than 1.46 Gy/day. At the highest dose rate of 21.8 Gy/day, no nauplii could mature, and died within 24 days. In blue mussel eggs, mortality reached 100% at 23-h postfertilization (h.p.f.) at a dose rate of 22 mGy/day (Hagger et al. 2005). This high mortality did not allow the determination of LDR_{50} at 72 h or later, but the authors give a 48-h LDR_{50} of 0.94 mGy/day. The EDR_{10} value estimated from their data is very low, i.e., 10^{-4} mGy/day. Chronic exposure of the polychaete *Ophryotrocha diadema*, from the egg to adult stage (11 weeks) at a single dose rate of 175 mGy/day, also led to a decrease in survival (18%) of organisms at the egg to larvae stage (Knowles and Greenwood 1997).

Tritium effects on invertebrate reproduction were studied in daphnids, polychaete worms, and brine shrimps. The resistant brine shrimp crustacean was the least sensitive among these organisms. However, reductions in the total number of nauplii, duration of breeding, broods and nauplii per brood, were observed from the lowest dose rates tested in these experiments, 290 mGy/day (Higuchi et al. 1980).

In the polychaete *O. diadema*, a 30% decrease in the mean number of larvae per worm was noted at a dose rate of 175 mGy/day from HTO, explained by a decrease in egg production and egg survival (Knowles and Greenwood 1997). Reproduction of worms was also studied for the same dose rate of γ rays. There was no significant difference between the β - or γ -irradiated groups for any endpoint, but results implied that the two groups may have affected different biological targets. The reason is that the reduced number of larvae resulted from a reduction in egg survival for tritium, whereas for γ -rays, reduced egg production was a more important effect.

The largest effects were observed in daphnids, whose fecundity was monitored over a five-generation exposure to HTO (Gudkov and Kipnis 1996). A decrease in several reproduction parameters was observed, such as the total number of young produced per female, the mean number of young in broods, and the number of broods during the life. At the maximum dose rate (39 mGy/day), there was no offspring produced in the entire experiment. The EDR_{50} values, estimated from these fecundity parameters, were 0.08 mGy/day (number of young in broods, number of broods in life) and 0.04 mGy/day (number of young produced during the life); the EDR_{10} value was 0.007 mGy/day.

3.1.3 Effects on Development

An effect of HTO exposure on the development of invertebrates was observed for low dose rates in four different studies, using brine shrimp, blue mussels, Pacific oysters, goose barnacles, and daphnids. The most resistant species to radiation was again the brine shrimp, which displayed a HNEDR as high as 21.8 Gy/day for hatchability of encysted dry eggs and for growth of nauplii. In Pacific oyster larvae, abnormalities were observed in larvae exposed for 48 h to a dose rate of 3 mGy/day (Nelson 1971). Although the effects seen were significant, the percentage of abnormalities remained low (11.4% vs. 5.1% in the control).

The freshwater microcrustacean, *Daphnia magna*, experienced the following several abnormalities during embryogenesis at a surprisingly low dose rate of 39 nGy/day: production of various sized eggs, uneven development of eggs, and dissolution of brooded eggs (Gudkov and Kipnis 1996). For example, at this dose, the maximum number of abnormalities (19.6% over all the generations) was observed and the proportion of abnormal developmental effects increased with each generation, reaching 37.5% at the fifth generation. It must be emphasized that a dose rate of 39 nGy/day is about two orders of magnitude lower than the normal background gamma dose rate (i.e., a few μ Gy/day), calling into question the validity of these data. The same trend was observed for the second dose rate of 39 μ Gy/day (mean of 19.1% abnormalities over all generations). This decrease was explained by a higher mortality of the weakened individuals and survival of more viable offspring.

Results expressed in terms of developmental abnormalities (all generations together) allowed the determination of an EDR_{10} of 0.04 mGy/day, and an EDR_{50} of 0.08 mGy/day. The blue mussel sensitivity appears comparable, since Hagger et al. (2005) observed only a few normal larvae after a 72-h exposure to a dose rate of 0.03 mGy/day (18% vs. 65% in controls), whereas for the other dose rates, no normal larvae were found (EDR_{10} of 0.017 mGy/day). Finally, one of the smallest LOEDR values recorded for the development of invertebrates exposed to tritium was in the goose barnacle in which negative effects on the molting index of poststage I larvae were observed at a LOEDR value of 15 μ Gy/day (Abbott and Mix 1979).

3.2 *Aquatic Vertebrates*

Most data on tritium effects in aquatic vertebrates were obtained on the fish medaka, in a series of experiments addressing the sensitivity of germ cells and early life stages. Data, albeit much less, were also acquired on rainbow trout and guppy (Table 2).

3.2.1 **Effects on DNA**

Despite the comprehensive studies performed on tritium effects to fish reproduction, there are few data on effects to DNA, and moreover, the latter were obtained at very high dose rates. A comparison was made of the cytogenetic effects to medaka eggs from fertilization (one-stage cell) to the blastula stage that was produced from exposure to tritium, ^{90}Sr - ^{90}Y , γ or X-rays (8 h.p.f.; Suyama et al. 1981). A dose-dependent increase of aberrant mitoses (i.e., frequency of cells with chromosomal bridges) was observed from the second studied dose rate (LOEDR of 555 mGy/day), at which the percentage of cells with chromosome bridges was twice that of the controls. The dose rate needed to obtain the same effect using γ rays was higher (LOEDR of 2340 mGy/day). From these the RBE can be estimated, viz., 4.2. Over the same range of dose rates, no effects were observed on hatchability and larval development.

Chromosome aberrations were also observed in microcultures of lymphocytes of the central mudminnow that was exposed to HTO or to γ irradiation (Suyama and Etoh 1985). Chromosome aberrations and SCE were observed from 23 mGy/day for HTO and from 64 mGy/day for γ rays. The RBE value was estimated by the authors to be 1.9, using the dose–response relationships for the HTO- and ^{137}Cs -induced aberration yields.

3.2.2 **Effects on Reproduction and Survival of Early Life Stages**

Mortality of fish eggs from exposure to low dose rates of tritium has been observed (Strand et al. 1972a). A significant increase in mortality (8.5%) was documented in rainbow trout eggs exposed for 3 weeks to HTO at a dose rate of 0.29 mGy/day. Similar sensitivity was seen for medaka embryos of the HO5 strain, viz., an EDR_{10}

Table 2 Effects induced in aquatic vertebrates by chronic tritium chronic exposure. Effect dose rate in mGy/day ($\mu\text{Gy/h}$)

Reference	Species	Dose rates	Exposure duration	Studied effect	Endpoint	EDR ₁₀ (mGy/day) ($\mu\text{Gy/h}$)	EDR ₃₀ (mGy/day) ($\mu\text{Gy/h}$)	LOEDR (mGy/day) ($\mu\text{Gy/h}$)	HNEDR (mGy/day) ($\mu\text{Gy/h}$)	RBE
Strand et al. (1972a)	<i>Salmo gairdneri</i> (rainbow trout, eggs)	0, 0.29, 2.9, 29.1 mGy/day	25 days	Mortality	Mortality after 25 days			0.29 (12.1)		
Strand et al. (1977)	<i>Salmo gairdneri</i> (rainbow trout, eggs)	0, 0.026, 0.23, 2.04, 20.1 mGy/day	20 days	Development immunity response	9 weeks after antigenic injection 11 weeks after antigenic injection			20 (833) 2 (83)		
Erickson (1971)	<i>Poecilia reticulata</i>	0, 73, 146, 291, 437, 582, 1455, 2910 mGy/day	17–30 days	Development	Higher proportion of ♂ Courtship frequency			73 (3,031) 73 (3,031)		
Hyodo-Taguchi and Egami (1977)	<i>Oryzias latipes</i> (medaka, adults)	0, 15, 29, 145, 291 mGy/day	30 days	Reproduction	Testes weight Number of primary spermatogonia Ib Germ cell survival	18 (764)	183 (7,625)	145 (6,040) 29 (1,200)		2.7 ^a
Etoh and Hyodo-Taguchi (1983)	<i>Oryzias latipes</i> (medaka, embryos-larvae)	0, 170, 340, 680, 850 mGy/day, 1.2 and 1.7 Gy/day ^a	10 days	Number of germ cell in embryos				170 (7,080)		
Hyodo-Taguchi and Etoh (1986)	<i>Oryzias latipes</i> (medaka, embryos to adults and F1)	0, 85, 170, 255, 340, 850 mGy/day, 1.7, 3.4 Gy/day ^a	10 days	Reproduction (fecundity and fertility of adults exposed at the embryo stage)	<i>Exposed</i> ♀, <i>control</i> ♂ Normal oviposition frequency Hatching rate	68 (2,830)	469 (19,461)	85 (3,542)		14.99 ^a 0.7 ^a
						201 (8,400)	642 (26,740)	340 (14,167)		3.8 ^a
						139 (5,812)	368 (15,322)	1,700 (70,800)		7.7 ^a
						33 (1,379)	250 (104,000)	255 (10,625)		0.2 ^a
						1,434 (59,800)	1,643 (68,500)	255 (10,625)		0.06 ^a
					<i>Control</i> ♀, <i>exposed</i> ♂	125 (5,225)	664 (27,670)	85 (3,542)		1.5 ^a
								340 (14,167)		0.7 ^a
						166 (6,917)	2,312 (96,350)	340 (14,167)		2.9 ^a

Hyodo-Taguchi and Etoh (1993)	<i>Oryzias latipes</i> (medaka, embryos)	0, 0.43, 0.85, 1.70 Gy/day ^a	10 days (from morula stage to hatching)	Vertebral malformations Mortality	Fertility (%)	169 (7,060) 45 (1,866)	169 (7,060) 318 (13,230)	340 (14,167) 340 (14,167)	4.3 ^a 0.2 ^a
Walden (1971)	<i>Gasterosteus aculeatus</i> (three-spined stickleback, eggs)	0, 1.455, 2.910, 5.821 mGy/day	Embryonic development	Development	Fry HO5 survival after 1 month	0.87 (36)	516 (21,503) 530 (22 × 10 ⁵)	340 (14,167) 430 (17,920)	2.7 ^a
Blaylock et al. (1971)	<i>Cyprinus carpio</i> (common carp)	0, 204, 1.455 mGy/day	72 h	Hatchability	Hatching			1.455 (60,400)	
Ichikawa and Suyama (1974)	<i>Fugu niphobles</i> (puffer)	0, 2.9 × 10 ⁻⁵ , 0.003, 0.29, 29.1, 2.910, 29,104 mGy/day	130 h	Development	Reduction of eye diameter			3 × 10 ⁴ (1.21 × 10 ⁶)	
	<i>Paralichthys olivaceus</i> (common flounder)	0, 2.9 × 10 ⁻⁵ , 0.003, 0.29, 29.1 mGy/day	92 h	Hatchability	Hatchability			3 × 10 ³ (1.21 × 10 ⁵)	
Suyama et al. (1981)	<i>Oryzias latipes</i> (Medaka eggs, blastula stage)	0, 111 and 555 mGy/day, 1.11, 5.55 and 11.1 Gy/day	8 h	Genotoxicity (chromosome aberrations)	Frequency of chromosome bridges hatchability and developmental abnormalities			29.1 (1.21 × 10 ³) 1,110 (46,250)	4.2 ^b
Suyama and Etoh (1985)	<i>U. limi</i> (central mudminnow lymphocytes)	0, 12, 23, 58, 116, 233 mGy/day	90 h chromosomal aberration 144 h SCE	Genotoxicity	chromosome aberrations, SCE			23 (972)	1.9 ^c

^aCalculated using EDR₁₀ values

^bCalculated using LOEDRs values

^cEstimated by the authors

of 0.87 mGy/day for survival at 1 month, when exposed from the morula stage to hatching (Hyodo-Taguchi and Etoh 1993).

Early exposure of medaka embryos (for 10 days) to HTO or to γ rays led to significant effects on subsequent fecundity and fertility as adults, after cross reproduction, i.e., exposed males or females were paired with control partners (Hyodo-Taguchi and Etoh 1986). No difference in sensitivity was observed between sexes, because fecundity was similar whether males or females were exposed. Hence, the EDR_{10} for oviposition frequency was 68 ± 62 mGy/day (mean \pm standard deviation) and 125 ± 86 (mean \pm standard deviation) mGy/day, respectively, for exposed females and males. However, differences appeared in terms of hatchability. For exposed females, eggs that were fertilized hatched normally in all groups, whereas for exposed males, the hatchability of eggs was affected at a dose rate as low as 340 mGy/day. This difference may derive from the lower repair ability of DNA damage in male germ cells than in female ones, and from residual tritium concentrations in important components of germ cells, from which regeneration of the surviving cells may be precluded. The fact that oviposition was also highly affected when only males were exposed indicates that an effect may have occurred on reproductive/courtship behavior, leading to a decrease of female egg laying. Several EDR_{10} values could be calculated for different endpoints, therefore allowing RBE values to be compared; these values ranged from 0.06 to 14.9 (Table 2).

Complementarily to the foregoing results, these authors studied the sensitivity of germ cells in medaka embryos that were exposed to tritium or to γ -rays until hatching, i.e., for 10–11 days (Etoh and Hyodo-Taguchi 1983). It seems that two populations of germ cells existed in the fry, a radiosensitive one and another that was radioresistant, since a decrease of germ cell number was observed until the third dose rate (340 mGy/day) was reached, whereupon the number of germ cells remained constant up to the highest dose rate studied (1.7 Gy/day). For radiosensitive germ cells, it was possible to calculate an EDR_{10} for germ cell survival of 18 mGy/day (EDR_{50} of 183 mGy/day). The EDR_{10} value estimated for γ irradiation was 48 mGy/day, which gave a RBE value of 2.7. For these radiosensitive cells, a 10-day LC_{50} of 195 mGy/day was reported for tritium (vs. 350 mGy/day for γ irradiation, the corresponding RBE value for which was 1.8).

Important effects were also observed on the fertility of adult medaka exposed to HTO for 30 days. Survival of primary spermatogonia Ib (the first stage after the stem cells) was affected by relatively low tritium concentrations, i.e., 29 mGy/day (Hyodo-Taguchi and Egami 1977). The corresponding EDR_{50} value was 50 mGy/day, at 10 and 30 days of exposure. Spermatogenesis was completely inhibited at the two highest dose rates, i.e., from 145 mGy/day. As a consequence, a decrease of testes weight was also observed from the same dose rate, reaching 40% after 30 days.

3.2.3 Effects on Development

Several malformations have been observed in fish exposed to tritium. In rainbow trout eggs, major malformations of the eyes and the body were observed

(Strand et al. 1972a). Oddly, significant effects on larvae were observed at the lowest dose (0.29 mGy/day), then decreased at higher dose rates, probably because the induced abnormalities resulted in death. Smaller eye size was also observed by Walden (1971) at high dose rates for the hatched fry of the freshwater fish, three-spine stickleback, and English sole (sea fish) exposed to HTO during their embryonic development. A 10% reduction in eye diameter was observed at the relatively high dose rates of 2.9 Gy/day, and a 20% reduction occurred at 29 Gy/day for both species. No effect was observed at the lowest dose rate tested (1.4 Gy/day). At high dose rates, Ichikawa and Suyama (1974) reported a 40% reduction in eye diameter for the puffer (*Fugu niphobles*), which had been exposed up to hatching to a single dose rate (29 Gy/day) of HTO. This reduced eye size and an observed swollen abdomen indicated that morphological development of those embryos had been retarded, resulting in smaller body size and a larger amount of remaining yolk. Vertebral malformations, such as fusion of three vertebrae, incomplete formation of vertebra or lack of a vertebral process, were also observed in medaka embryos that were exposed from the morula to hatching stages; the EDR_{10} for this effect was 798 mGy/day (Hyodo-Taguchi and Etoh 1993). The effects on development of the primary immune response were studied in rainbow trout exposed as embryo-larvae for 20 days to tritium (Strand et al. 1977). A 50% decrease of agglutinine synthesis, an antibody induced in response to vaccination, was reported as an effect at the highest dose rate (20 mGy/day) 9 weeks after exposure, and at 2 mGy/day 11 weeks after exposure.

Using a hatchability endpoint, Blaylock et al. (1971) found no significant differences between control carp eggs and those that had been exposed to 204 and 1,450 mGy/day of HTO. Ichikawa and Suyama (1974) also found no effect of tritium dose rates up to 29 mGy/day on the hatching of flounder eggs, but a small reduction of hatching occurred for puffer (*Fugu niphobles*) eggs at a dose rate of 2910 mGy/day.

In guppies, tritiated water produced a significant effect on the sex ratio, with an increased proportion of males occurring at 73 mGy/day. A dose-dependent decrease of courtship behavior was also observed, together with a decrease in the rate of development of male characteristics (Erickson 1971).

4 Review of Effects from External Gamma Irradiation

4.1 Aquatic Invertebrates

4.1.1 Effects on DNA

As occurred with studies on tritium, although DNA is known to be the primary target for ionizing radiation, few studies have addressed the genotoxicity in invertebrates induced by γ irradiation (Table 3). A few studies have been conducted using the polychaete worm, *Neanthes arenaceodentata*, in which a doubling in the number

Table 3 Effects induced in aquatic invertebrates by chronic external gamma-irradiation exposure. Effect dose rate in mGy/day ($\mu\text{Gy/h}$)

Reference	Species	Dose rates	Exposure duration	Studied effect	Endpoint	EDR ₁₀ (mGy/day) ($\mu\text{Gy/h}$)	EDR ₅₀ (mGy/day) ($\mu\text{Gy/h}$)	LOEDR (mGy/day) ($\mu\text{Gy/h}$)	HNEDR (mGy/day) ($\mu\text{Gy/h}$)
Pesch and Young (1981)	<i>N. arenaceodentata</i> (marine polychaete worm, larvae)	0, 72 mGy/day 0, 144 mGy/day	24 days 48 days	Genotoxicity	Chromosomal aberrations			72 (3,000)	
Harrison and Rice (1981)	<i>N. arenaceodentata</i> (marine polychaete worm, larvae)	0, 9, 6, 24, 120, 144, 300, 480, 600, 1,680, 3,120 mGy/day	12 and 24 h	Genotoxicity Development	Sister chromatid exchange Abnormal larvae 6 and 17 days postirradiation			9.6 (400)	2,760 (11.5×10^4)
Knowles and Greenwood (1994)	<i>O. diadema</i> (marine polychaete worm)	0, 41, 77, 185, 329 mGy/day	1 year (seven generations)	Reproductive performance	Number of larvae per worm, second generation	18.0 (750.5)	136 (5,685)	77 (3,208)	
					Survival of larvae, second generation, day 62	0.86 (35.8)	89 (3,726)	173 (7,200)	
					Survival of larvae, third generation, day 62	56.6 (2,360)			329 (13.7×10^3)
Knowles and Greenwood (1997)	<i>O. diadema</i> (marine polychaete worm, adults F0 and F1)	0, 175 mGy/day	11 weeks (one generation)	Reproductive performance	Growth, time to sexual maturity Number of egg sacs, eggs and larvae produced			175 (7,292)	
Harrison and Anderson (1988)	<i>N. arenaceodentata</i> (marine polychaete worm, eggs to adults and F1)	0, 4, 6, 50.4, 408 mGy/day	130 days 12-days exposure during embryogenesis	Reproductive performance Embryo survival	Embryo survival Egg number Embryo survival			4.6 (190)	175 (7,292)
								408 (17×10^3)	408 (17×10^3)

Gilbin et al. (2008)	<i>Daphnia magna</i> (daphnid)	0, 9, 97, 648 mGy/day	23 days (five broods)	Survival, growth Reproductive performance	Survival, growth Reduced brood size, advance in release of broods Larval survival to 5 days of starvation, brood 1	648 (27×10^3) 648 (27×10^3)	648 (27×10^3)
Baptist et al. (1976)	<i>M. mercenaria</i> (marine hard clam) <i>A. irradians</i> (Atlantic scallop)	0, 1.4–1.7, 14–22, 216–240, 3,840–8,880 mGy/day	14 months 3 months	Survival Growth Survival	Survival of juveniles Growth Survival of juveniles	1,188 (49.5×10^3) ^a 347 (14.5×10^3) ^a	8,880 (37×10^4)
Cooley and Miller (1971)	<i>Physa heterostropha</i> (freshwater snail aged of 45 days)	0, 240, 2400, 6000 mGy/day	Entire life span	Survival	Survival, egg hatchability, growth	2,400 (10^5)	
Marshall (1962)	<i>Daphnia pulex</i> (daphnid)	0, 5308, 11,151, 12,152, 15,714, 17,670 mGy/day	35 days (six broods)	Reproductive performance Survival	Number of eggs per snail Life-span, survival	1,340 (55.8×10^3) ^a 17,670 (73.6×10^4)	
Marshall (1966)	<i>D. pulex</i> (daphnid)	0, 5308, 11151, 12152, 15714, 17670 mGy/day	55 weeks	Reproductive performance Survival Reproductive performance	Population birth rate, per day population rate of increase, "r" Number of aborted embryos	4,360 (18.2×10^4) 1,080 (45×10^3)	

^aEDR₁₀ value calculated in Garnier-Laplace et al. (2010)

of SCEs was observed in larvae, following exposure to 9.6 mGy/day (Harrison and Rice 1981). Worms exposed to higher dose rates (1,680 and 3,120 mGy/day) had lower SCEs than did the controls, probably indicating that apoptosis had occurred. There were no exposure effects at the individual level, since no difference in the number of abnormal larvae or survival rates was observed in larvae 6 and 17 days after irradiation at dose rates of 528 and 2,760 mGy/day, or in another study, at 408 mGy/day (Harrison and Anderson 1988). However, a substantial increase of larvae mortality and egg number was observed when parents were exposed, which may derive from chromosomal aberrations having caused cell death and mutations in gametes. Comparing embryonic survivorship data obtained from chronic vs. acute exposure reveals that DNA repair may not be efficient in gametes of this species. As a result of *N. arenaceodentata* having long synchronous periods of gametogenesis, this organism may be more vulnerable to cumulative effects of exposure to γ rays.

4.1.2 Effects on Survival of Early Life Stages and Reproduction

Reproductive effects from γ irradiation were documented to occur for the polychaete *O. diadema*, when exposed for seven generations (over 1 year of exposure) to low dose rates (Knowles and Greenwood 1994). In generation 1, several reproductive parameters were decreased (number of egg sacs, eggs and larvae produced), but only at the highest dose rate (329 mGy/day); in generations 2 and 3, all parameters (number of egg sacs, eggs and larvae produced, survival of egg to larvae) were decreased in a dose-dependent manner for all dose rates. The EDR_{10} value for larvae production in the second generation was estimated to be 18 mGy/day. In the seventh generation, a clear recovery was seen. Survival of eggs to larval stages was affected slightly in the first generation at the highest dose rate, and in a dose-dependent manner for generations 2 and 3. No effect was observed for generation 7. Mortality also increased for older worms (62 days) in the second generation ($EDR_{10}=0.86$ mGy/day) and in the third one ($EDR_{10}=56.6$ mGy/day) to a lesser extent, whereas a recovery was seen in the seventh generation. The reasons for these differences must lie in the fact that worms in generation 1 were exposed first as free living larvae, whereas in generations 2 and 3, the organisms had been subjected to radiation from the fertilization stage, when irradiated gametes came together. The question as to whether a selection of the more resistant individuals occurred at generation 7 must still be addressed. No radiation effect on growth rate or time to reach sexual maturity was observed at the dose rates studied (from 41 to 329 mGy/day).

The same trend (reduction in egg sacs, eggs, and larvae) was observed in a similar study (Knowles and Greenwood 1997), in which organisms of *O. diadema* were exposed to a single dose rate of 175 mGy/day for 11 weeks (from the egg prior to its being laid to when the worms were approaching the end of their lives).

The influence of the exposure period on the reproductive performance of another polychaete worm, *N. arenaceodentata*, was studied at similar dose rates (Harrison and Anderson 1988). Irradiation carried out during embryogenesis only (from

spawning to hatching of larvae, i.e., 12 days) did not affect embryo survival, even at a dose of 408 mGy/day. However, life-time irradiation of parent worms from their first being spawned, caused a significant reduction in the survival of embryo offspring, even at a dose rate of only 4.6 mGy/day. A significant reduction in egg number was observed, but only at the highest dose rate.

These results show that *N. arenaceodentata* and *O. diadema* exhibited different sensitivities that were dependent on the endpoint being measured. Hence, embryonic survival may be more radiosensitive than egg production in *N. arenaceodentata*, and the reverse for *O. diadema*.

The life history strategy of an organism can also strongly influence results. *N. arenaceodentata*, which has long and synchronous periods of gametogenesis, may be more vulnerable to the cumulative effects of chronic exposure to genotoxic substances.

Species sensitivity was also drastically different in two other marine species, *Mercenaria mercenaria* and *Argopecten irradians*, that were exposed to increasing dose rates for 3–14 months (Baptist et al. 1976). Although *M. mercenaria* survival was affected after 159 days of exposure (EDR₁₀ of 1188 mGy/day), no significant effect was observed on juvenile scallops after 84 days of exposure up to doses of ca. 9,000 mGy/day.

For the freshwater crustacean *D. magna*, reproduction was also affected by exposure to γ irradiation at levels of <24 h.p.f. for 23 days (Gilbin et al. 2008). The fecundity rate was significantly affected at 650 mGy/day, with early release and reduced size of broods. As a result, the intrinsic rate of natural population increase (“*r*”), defined as a function of survival and fecundity, decreased by 21% for the first generation exposed to the highest dose rate. Furthermore, the neonates produced were less resistant to starvation (EDR₁₀ of 403 mGy/day). In another daphnid species, *Daphnia pulex*, that was also exposed for one generation (35 days), but to much higher dose rates, Marshall (1962) showed that there was a negative correlation between fertility and dose rate; the corresponding EDR₁₀ was 6663 mGy/day. As for *D. magna*, this reduction in fertility resulted from reduced fecundity rather than from increased prenatal mortality of embryos. The intrinsic growth rate of natural population increase was reduced as a linear function of the square of dose rate, and equaled zero at a dose rate of 16,300 mGy/day. This almost entirely resulted from a decline in birth rate at doses exceeding 7,000 mGy/day, whereas mortality increased only at dose rates exceeding 14,000 mGy/day. A 20% decrease of “*r*” was observed at 7,000 mGy/day vs. 648 mGy/day in the Gilbin et al. (2008) study. *D. pulex* and *D. magna* may have different sensitivities to irradiation, but these differences may also derive from different experimental conditions, e.g., such as food or experimental conditions.

The effect of γ -rays on population dynamics of *D. pulex* was also studied over several generations (for 55 weeks), under conditions of intraspecific competition for food (Marshall 1966). These exposure conditions led to population extinction at a dose rate of 4,360 mGy/day, which was lower than the value of 16,300 mGy/day found in the previous study (Marshall 1962). In contrast to the 1962 study, brood size increased with increasing dose rate, because the reduction of fecundity was

indirectly compensated for by the increase in food supply per individual. Finally, because the individuals in the Marshall (1966) study were continuously exposed from the earliest embryonic development, their life-span shortening was much more significant than in the first study, in which exposure started at birth.

A freshwater snail species, *Physa heterostropha*, was studied by being exposed from the age of 45 days for their entire life span; results were a significant decrease in snail survival and reproductive performance at a dose rate as low as 1.34 Gy/day (EDR₁₀) (Cooley and Miller 1971). The highest dose rate (6,000 mGy/day) produced extinction of the population in one generation. Adaptation of this species to a low level chronic contamination (6.5 mGy/day) was studied by comparing fecundity levels of the irradiated population with a control population in both field and laboratory (Cooley 1973). When the frequency of egg capsule production was reduced in the irradiated population, the eggs per capsule increased, resulting in a compensation mechanism. The same tendency was seen for the fish species *Gambusia affinis* (increased brood size in exposed field populations; Blaylock 1969).

4.2 Aquatic Vertebrates

4.2.1 Effects on DNA

There are more data on DNA damage induced by γ irradiation in aquatic vertebrates (Table 4) than for invertebrates. Primary lesions of DNA, such as strand breaks (determined using the Comet assay), were measured in zebrafish cells exposed in vitro (primary cultures) for 24 h to external ¹³⁷Cs γ rays. An increased sensitivity of male germ cells was seen as compared to hepatocytes (Adam et al. 2006), with a LOEDR for DNA alterations in sperm cells of 1 mGy/day vs. 750 mGy/day for hepatocytes. A dose-dependent increase of DNA double strand breaks (DBSs) and micronuclei was also observed from 10 mGy/day in ZF4 cells (embryonic fibroblasts; Pereira et al. 2011). The same sensitivity was observed in vivo on fertilized eggs exposed to external γ irradiation for 1 and 2 days, with an increase of DNA damage observed from a dose of 1 mGy/day (Bourrachot 2009). For 2-day-old larvae of the same species (i.e., 5-6 days postfertilization) that were exposed to external ¹³⁷Cs γ irradiation at dose rates ranging from 9.6 to 178 mGy/day, genotoxic effects also occurred at doses as low as 29 mGy/day (measured by using the Comet assay; Jarvis and Knowles 2003).

For comparable dose rates, no genotoxicity was observed in a marine fish species, the plaice, that were exposed to 6 to 24 mGy/day, for 64 and 167 days (Knowles 1999). As suggested by the authors, it is probable that the methods used (micronuclei counts and flow cytometry) may not have been sensitive enough to detect an effect. The chosen life stage (adults) and cell type (erythrocytes) may also have been less sensitive than early life stages and germ cells.

In another freshwater fish species, the medaka, whose eggs were exposed for 8 h to a range of high dose rates, chromosome bridges were observed at the lowest dose rate studied (2340 mGy/day) (Suyama et al. 1981).

Table 4 Effects induced in aquatic vertebrates by chronic external gamma-irradiation exposure. Effect dose rate in mGy/day ($\mu\text{Gy/h}$)

Reference	Species	Dose rates	Exposure duration	Studied effect	Endpoint	EDR ₁₀ (mGy/day) ($\mu\text{Gy/h}$)	EDR ₅₀ (mGy/day) ($\mu\text{Gy/h}$)	LOEDR (mGy/day) ($\mu\text{Gy/h}$)	HNEDR (mGy/day) ($\mu\text{Gy/h}$)
Tsyusko et al. (2007)	<i>Oryzias latipes</i> (medaka, parents)	0, 68 mGy/day	45 days for females 153 for males	Genomic instability	Mutation frequencies at tandem repeat loci			68 (2,833)	
Woodhead (1977)	<i>Poecilia reticulata</i> (guppy)	0, 40, 96, 305 mGy/day	920 days	Reproductive output, histology	Eggs laid, spawning events, gonad histology, onset of infertility Lifetime fecundity	2.5 (105.5) ^a 12.4 (516) ^a		40 (1,667)	
Knowles (1999)	<i>Pleuronectes platessa</i> (plaice, adult male erythrocytes)	0, 6, 12, 24 mGy/day	197 days	Reproduction (testes histology) Genotoxicity	F1 survival, sex ratio Mean proportion of testes occupied by sperm cells Mean proportion of testes occupied by non germinal cells ♂ GSI	1.13 (47) ^a 0.014 (0.6) ^a 499 (20,792) ^a			305 (12,708) 24 (1,000)
Etoh and Hyodo-Taguchi (1983)	<i>Oryzias latipes</i> (medaka, embryonic larvae)	0, 114, 238, 364, 476, 1,080, 2,120 mGy/day	10 days	Number of germ cell in embryos exposed from 2 h.p.f to 10 days	Germ cell survival	48.3 (2,012)			
Hyodo-Taguchi et al. (1982)	<i>Oryzias latipes</i> (medaka, adults)	0, 29, 68, 156, 242, 843 mGy/day	120 days	Reproduction	Testis weight after 30-day exposure GSI after 120 days Number of primary spermatogonia Ib at 10 days			156 (6,040) 29 (1,210) 29 (1,210)	
Hershberger et al. (1978)	<i>Oncorhynchus ishawytscha</i> (Chinook salmon)	0, 5.24, 12.6, 27.1, 48.5, 97, 194, 165–485 mGy/day	67–86 days	Development	Return to freshwater as adults, growth, F1 mortality, age at return and male sterility			97 (4,042)	
Knowles (1992)	<i>Oncorhynchus mykiss</i> (Rainbow trout)	0, 24, 46, 112 mGy/day	246 days	Development (humoral immune response)	Antibody titre			112 (4,667)	

(continued)

Table 4 (continued)

Reference	Species	Dose rates	Exposure duration	Studied effect	Endpoint	EDR ₁₀ (mGy/day) (μ Gy/h)	EDR ₃₀ (mGy/day) (μ Gy/h)	LOEDR (mGy/day) (μ Gy/h)	HNE DR (mGy/day) (μ Gy/h)
Bonham and Donaldson (1972)	<i>Oncorhynchus tshawytscha</i>	0, 4.4, 11, 24, 43, 87, 174, 435 mGy/day	80 days	Development	Gonadal development	*49 (2,046)			
Adam et al. (2006)	<i>Danio rerio</i> (zebrafish, primary cell culture)	0, 1, 10, 100, 750 mGy/day	24 h	Genotoxicity	DNA damage in male germ cells Hepatocytes			1 (40) 750 (31,250) 10 (400)	
Pereira et al. (2011)	<i>Danio rerio</i> (zebrafish, ZF4 cells)	0, 10, 100, 750 mGy/day	24 h	Genotoxicity	H2AX, DNApk, micronuclei			29 (1,200)	
Jarvis and Knowles (2003)	<i>Danio rerio</i> (zebrafish, larvae 2 days posthatching)	0, 9.6, 29, 173 mGy/day	1 and 24 h	Genotoxicity	DNA damage in larvae of 2 d				
Bourrachot (2009)	<i>Danio rerio</i> (zebrafish, eggs 1–3 h.p.f.)	0, 1, 10, 100, 750 mGy/day	15 days	Development Survival Genotoxicity	Hatching Mortality at 13 d.p.f. DNA damage in 24 and 48 h.p.f. embryos			10 (400) 100 (4,000) 1 (40)	
Knowles (2002)	<i>Danio rerio</i> (zebrafish, adults, 22 weeks old)	0, 7.2, 24, 178 mGy/day	Ca. 30 weeks	Reproductive output, histology	Eggs laid, spawning events, egg viability, testes histology			178 (7,417)	
Hinton et al. (2004)	<i>Oryzias latipes</i> (medaka, adults)	0, 350 mGy/day	28 days	Reproduction	Egg laid, egg viability and hatchability			350 (14,583)	
Rackham and Woodhead (1984)	<i>Ameba splendens</i> (Ameba)	0, 175 mGy/day	95 days	Reproduction (testes histology)	Complete reduction in all stages of germ cells except the primary spermatogonia; increase of eggs degenerating			175 (7,300)	
Egami and Hama-Furukawa (1981)	<i>Oryzias latipes</i> (medaka, embryos to adults)	0, 272, 514 mGy/day, 1.26, 2.33, 5.24 Gy/day	50 days	Reproduction (testes histology, survival)	♂ germ cell number ♀ germ cell number Gonad absence Mortality of fry			272 (11,333) 514 (21,417) 1,260 (52,500)	
					♂ GSI ♀ GSI				2,111 (87,960) ^a 590 (24,584) ^a 800 (33,361) ^a

Hyodo-Taguchi and Etoh (1986)	<i>Oryzias latipes</i> (medaka, embryos to adults and F1)	0, 61, 105, 240, 910 mGy/day, 1.4, 2.5 Gy/day	10 days	Reproduction (fecundity and fertility of adults exposed at the embryo stage)	Exposed ♀, control ♂ Normal oviposition frequency Hatching rate Number of complete infertile ♀ Total oviposition frequency Number of fertilized eggs per oviposition Fertility (%) Control ♀, exposed ♂ Normal oviposition frequency Hatching rate Number of complete infertile ♂ Total oviposition frequency Number of fertilized eggs per oviposition Fertility (%) Chromosome bridges	1,014 (42,250) 2,080 (86,800) 2.5 × 10 ³ (1 × 10 ⁶) 2.5 × 10 ³ (1 × 10 ⁶) 240 (10,000) 2.5 × 10 ³ (1 × 10 ⁶) 2.5 × 10 ³ (1 × 10 ⁶) 2.5 × 10 ³ (1 × 10 ⁶) 88 (3,700) 1,643 (68,500) 1,400 (58,300) 193 (8,050) 1,386 (57,800) 240 (10,000) 240 (10,000) 485 (20,200) 2,312 (96,350) 1,400 (58,300) 733 (30,500) 1,024 (42,700) 1,400 (58,300) 9 (386) 1,260 (52,500) 105 (4,375) 342 (14,200) 105 (4,375) 2,340 (97,500)
Suyama et al. (1981)	<i>Oryzias latipes</i> (medaka, eggs)	0, 2.3; 4.4; 7.9; 11.3; 17.7 Gy/day	8 h	Genotoxicity (chromosomal aberrations)		
Shimada and Shima (2001)	<i>Oryzias latipes</i> (medaka, sperm and spermatids)	0, 2.40, 4.75, 7.10, 9.50 Gy (at a dose rate of 0.95 Gy/day)	0, 2.5, 5, 7.5, 10 min	Genomic instability	Mosaic and whole-body mutations in <i>wf</i> locus F1	2.4 × 10 ^{3b}
Shimada and Shima (2004)	<i>Oryzias latipes</i> (medaka, spermatogonial stem cells)	0, 4.75 Gy (dose rate of 0.95 Gy/day)	0, 5 min	Genomic instability	Mosaic and whole-body mutations in <i>wf</i> locus F1	4.75 × 10 ^{3c}
Shimada et al. (2005)	<i>Oryzias latipes</i> (medaka, spermatogonial stem cells)	0, 1.9, 3.2, 4.75 Gy (dose rates of 0.3 mGy/day and 0.95 Gy/day)	0, 4.4, 7.4, 11 days 0, 2, 3.4, 5 min	Genomic instability	Specific locus mutations at five pigmentation loci	1.9 Gy (at dose rates of 0.3 mGy/day or 0.95 Gy/day) ^b

^aEDR₁₀ value calculated in Garnier-Laplace et al. (2010)

^bLOED: lowest observed effect dose

^cHNEI: highest no effect dose

Mutations were also detected in fish exposed to acute doses of gamma rays (from 2.4 to 9.5 Gy at 0.95 Gy/min) (Shimada and Shima 2001). These genetic alterations appeared either as whole-body mutants or mosaic mutants (i.e., only some cells mutated), the frequency of the latter being ca. four times higher than the first. Moreover, most of the mosaic mutants arising from paternal irradiation appeared to die from developmental abnormalities (e.g., small body, weak heartbeat, or slow blood circulation). Using different crosses of wild-type medaka and *wl* mutants, it was shown that whole-body mutants were produced from genetic alterations that occurred at the gamete stage, whereas mosaic mutants were produced from genetic instability at or after the two-cell stage. No increase of mutations was observed when stem spermatogonia were irradiated (Shimada and Shima 2004). This specific-locus test was also applied to compare the same dose, delivered at a “low” dose (432 mGy/day) or at the same high dose as described above (0.95 Gy/min). Both dose rates induced a significant increase of mutation frequencies, which resulted in the major portion of mutant embryos being not viable. The mutation frequency was twice to four fold higher at the highest dose rate.

Radiation-induced untargeted germline mutations were also observed in medaka following chronic exposure (dose rate of 68 mGy/day for 45–153 days) (Tsyusko et al. 2007). The microsatellite mutation rate was higher in the offspring from exposed parents than from the control parents. Furthermore, the magnitude of the mutational response was greater than expected by direct DNA damage, suggesting that indirect mechanisms, remote in time and space, contributed to the mutations.

4.2.2 Effects on Reproduction and Survival of Early Life Stages

The effects of chronic gamma exposure at a range of low dose rates (from 7.2 to 178 mGy/day) were studied in zebrafish over a period of more than 30 weeks (Knowles 2002). A significant decrease in the mean number of eggs per spawning opportunity was reported at the highest dose rate (41 eggs in control vs. 5.8 in the 178 mGy/day group, i.e., a decrease of 86%). This decrease primarily resulted from reduced spawning events, since nearly all pairs had ceased laying eggs from week 20 (i.e., a decreased spawning events). Though smaller, a decrease in the number of eggs per spawning event was also observed (61.8 in control vs. 41.3 in the 178 mGy/day group, i.e., decrease of 33%). The viability of these eggs was also affected (81% in the control vs. 41% in the 178 mGy/day group). A small, but not significant decrease in the mean number of eggs hatched was also observed. In addition, fish from the group exposed to the highest dose rate (which had ceased producing eggs) were paired with unirradiated partners for 5–6 weeks. A few irradiated females produced eggs, but none were fertile. However, many of the irradiated males did couple with unirradiated females to produce viable eggs; fertility varied between 21% and 75%. The fact that eggs were laid from irradiated females that had not laid eggs for 20 weeks suggests that some stimulation to lay eggs was provided by the presence of unirradiated fish, but not irradiated ones. These findings on reproductive output were supported by histological effects in the highest exposure group (178 mGy/day).

Indeed, several testes contained no spermatogenic tissue, and retained only the outer lining and cyst wall structures. In contrast, only moderate changes were observed in the ovaries in the fish of this group; the changes noted were small areas of atresia associated with a mass of stromal tissue.

These results are in agreement with those obtained in another tropical fish species, the medaka, in which a reduction in egg number, egg viability, and hatchability was observed at a dose rate of 350 mGy/day, following 28 days of irradiation (Hinton et al. 2004). A third study in tropical fish, the guppy, was also performed. The total life-time breeding performance was studied for 920 days in guppies exposed to dose rates of 40, 96 and 305 mGy/day (Woodhead 1977). The mean life-time fecundity was affected at doses as low as 12.4 mGy/day (EDR_{10}), and occurred because of a reduction both in the number of broods and of brood size, the former being the probable main factor. Additionally, the mean time to the initial onset of infertility became shorter with increasing dose rates (660 days for the control vs. ~300 days for 40 and 96 mGy/day, and 100 days for 305 mGy/day). This apparent sterility was often linked with abnormalities in gonad histology. For 2–3 individuals exposed to the lower dose rate, and all fish exposed to the highest dose rate, both male and female gonads were devoid of germ cells. The mean survival of neonates, survival to maturity, and sex ratio were not affected.

These results are supported by other studies at the tissue or cell level. In medaka fry exposed from fertilization until 50 days of age, a reduction in germ cells was observed to occur in a dose-dependent manner (Egami and Hama-Furukawa 1981). The degeneration of male germ cells occurred at dose rates as low as 272 mGy/day (~50% of reduction after 30 days), and was complete following exposure to 1.26 Gy/day. In females, the entrance of oocytes into meiotic prophase was retarded at a dose rate of 514 mGy/day. Fish exposed to 1.26 and 2.33 Gy/day during the embryonic phase developed into adults having normal secondary sexual characteristics, but their germ cells were completely destroyed. Hence, it seems that a regeneration of germ cells is possible for dose rates lower or equal to 514 mGy/day. The gonad weight was normal up to 514 mGy/day, whereas it was null at higher dose rates.

In male medaka exposed as adults, a transient reduction in the gonadosomatic index (GSI) was observed at a dose rate of 29 mGy/day, whereas a dose-dependent reduction was seen at dose rates of 68–843 mGy/day for 30 days, which was maintained for 120 days (Hyodo-Taguchi et al. 1982). Spermatogenesis was almost completely inhibited following 30 days of exposure to 843 mGy/day (decreased number of primary spermatogonia Ib). At 10 days of exposure, primary spermatogonia Ib survival was affected from the lowest studied dose rate (29 mGy/day).

At a similar dose rate (175 mGy/day), spermatogenesis of adult *Ameba splendens* was disrupted after 5 days and completely inhibited after 52 days, at which time there was a complete loss of primary and secondary spermatocytes and spermatids and no sperm production. The effect of radiation was more pronounced on the production of secondary spermatogonia, i.e., on the mitosis of the stem cells, which themselves did not completely disappear until day 95. The time needed for recovery of these effects increased as the exposure duration lengthened (i.e., a recovery of 85–90% after 125 days for fish exposed for 21 days, and 5–10% for

those exposed for 40 days). As regards ovaries, the primordial oocytes had completely disappeared by day 105, and many of the eggs obtained showed signs of degeneration.

A higher radiosensitivity was observed for plaice, whose testis weight and sperm content were reduced by approximately 50% from those of controls after 197 days of exposure at only 5.8 mGy/day (Knowles 1999). In this experiment, the existence of a high spermatocyte to sperm ratio suggested that the effect responsible for sperm reduction occurred at the spermatocyte to sperm transition, rather than from damage to spermatogonia. As described above in Sect. 3, Etoh and Hyodo-Taguchi (1983) studied the sensitivity of germ cells in medaka embryos exposed to γ -rays or to tritium, or until hatching, i.e., for 10–11 days. The EDR_{10} value for germ cell survival was 48.3 mGy/day, although apparently two populations of cells of different radiosensitivity existed. For the more radiosensitive germ cell type, it was possible to calculate a 10-day LC_{50} of 350 mGy/day for γ irradiation.

In a second study, the effects of these early radiation-induced germ cell losses on lifetime reproduction were studied (Hyodo-Taguchi and Etoh 1986). As in the first experiment described above, embryos were exposed from the morula stage until hatching (10 days) to a range of doses of tritium or γ -rays. Fecundity and fertility were then determined after 4–8 months. Mating pairs consisted of irradiated males and control females, or the reverse as described in Sect. 3. For γ -ray-exposed fish, the number of eggs per oviposition were affected only at high dose rates (EDR_{10} of 1,074 and 733 mGy/day, respectively, for irradiated females and males). The number of fertilized eggs was impacted at lower dose rates (EDR_{10} of 33 and 45 mGy/day, respectively, in exposed females and males). The hatchability was affected at high dose rate for females (LOEDR of 2.5 Gy/day) and at a much lower dose rate for males (LOEDR of 240 mGy/day).

4.2.3 Effects on Development

The effect of chronic γ irradiation on the development of the Chinook salmon was studied at low dose rates (Hershberger et al. 1978). Eggs and alevins were exposed for 90 days to dose rates of 5.24–485 mGy/day, and different biological effects were studied when fish migrated back to the pond as adults. The effects that were observed at dose rates as low as 97 mGy/day included a decreased number of fish returning to spawn, retardation of growth, increased mortality of small salmon in freshwater, increased age at return, and apparent sterility of males. A retardation of gonadal development in smolts from the same treatment (EDR_{10} of 49 mGy/day) was also observed (Bonham and Donaldson 1972). Vertebral malformations such as fusion of three vertebrae, incomplete formation of vertebra or lack of vertebral process also occurred in medaka embryos from the morula stage to hatching, when exposed to γ -irradiation at levels as low as 430 mGy/day (Hyodo-Taguchi and Etoh 1993); comparable results also occurred from tritium exposure (RBE of 1).

An acceleration of hatching was observed in zebrafish eggs at dose rates of 10 and 1,000 mGy/day, while no significant difference was seen at 100 mGy/day

(Bourrachot 2009). Larval mortality increased greatly after hatching, reaching almost 100% for larvae exposed to 100 and 1,000 mGy/day at 13 days postfertilization (vs. 26% in controls). The only malformation observed was edemas of pericardial cavity, at 75 h.p.f. at 1,000 mGy/day.

The effect of chronic irradiation for 246 days on the humoral immune response in rainbow trout was studied for a range of low dose rates (Knowles 1992). The antibody response decreased with increasing dose rates, and was significantly lower in trout receiving the highest dose rate (110 mGy/day).

5 Discussion

5.1 *Comparison of Tritium and External Gamma-Irradiation Effects*

5.1.1 Degree to which the Benchmark Value Protects

Tritium effects were studied over a large range of dose rates in invertebrates from 39 nGy/day (Gudkov and Kipnis 1996) to 22 Gy/day (Higuchi et al. 1980), and in vertebrates from 29 nGy/day to 29 Gy/day (Ichikawa and Suyama 1974). Combined, the results suggest that the dose rates of tritium induce effects in invertebrates at quite low levels as compared to gamma external irradiation. Indeed, except for the two studies conducted at rather high dose rates (Higuchi et al. 1980; Knowles and Greenwood 1997), the EDRs for tritium generally ranged between 0.03 and 0.3 mGy/day for all endpoints studied (genotoxicity, abnormalities, fecundity, or embryo-larvae mortality). These values are less than 0.24 mGy/day, which is the previous level or benchmark recommended to protect aquatic ecosystems from external γ irradiation (Garnier-Laplace et al. 2006). In addition, several EDR_{10} values calculated for tritium (e.g., 0.2 μ Gy/day for chromosome aberrations in blue mussels or 0.8 μ Gy/day in goose barnacles for molting parameters) are in the range of a few μ Gy/day or even lower, i.e., below one order of magnitude of background γ radiation.

For vertebrates, the dose rates at which effects appear are higher than for invertebrates and exceed the benchmark value. Our review indicates that effects on vertebrates appear at dose rates ranging from approximately several tens to several hundreds of mGy/day. The research results of Strand et al. (1972b, 1977) are exceptions in that they found mortality and development effects in rainbow trout at the lower dose rates of 0.29 and 2 mGy/day.

For external γ irradiation, the dose rates used for invertebrate studies ranged from 1.4 mGy/day (Baptist et al. 1976) to 17.7 Gy/day (Marshall 1966), and for vertebrates from 1 mGy/day to 5.24 Gy/day. Contrary to the tritium data on invertebrates, the data derived from invertebrates exposed to external γ irradiation did not exceed the recommended benchmark value thought to be protective of ecosystems

(0.24 mGy/day; Garnier-Laplace et al. 2006). Indeed, the lowest value for significant effects in invertebrates was observed for the polychaete worm *N. arenaceodentata*, whose survival of embryos born from exposed parents was affected at 4.6 mGy/day (Harrison and Anderson 1988). Genotoxicity was also seen in larvae exposed to 9.6 mGy/day from spawning, but their survival was not affected (Harrison and Rice 1981), which means that DNA damage was repaired or was not lethal. In vertebrates, the lowest value for a significant effect was observed in zebrafish at 1 mGy/day. The observed effect was genotoxicity, induced either to primary cells (male gametes) or in embryos aged of 24 or 48 h.p.f. (Adam et al. 2006; Bourrachot 2009). Effects on testes were also seen at low dose rates (9 mGy/day) for plaice (Knowles 2002) and at 29 mGy/day for medaka (Hyodo-Taguchi et al. 1982). When the endpoint measured was reproductive output, significant effects were seen for the guppy at 40 mGy/day (Woodhead 1977), and for zebrafish at 178 mGy/day (Knowles 2002). Acute transgenerational genomic instability was evaluated in several studies, but chronic exposure was addressed in only one study; the results of this study indicated significant genomic instability at 68 mGy/day (Tsyusko et al. 2007).

5.1.2 RBE and Dose Calculation

The effect of an absorbed dose depends on the type and energy of the irradiation. The type of irradiation and its energy is taken into account in radiation biology by using an RBE factor that normalizes the relative effectiveness of different radiation types to produce the same biological effect.

The results presented in this chapter suggest that RBEs are highly variable, and most importantly, are endpoint-dependent. As a result, RBEs ranged from 0.06 to 14.9, depending on the biological effect studied (Tables 1 and 2). The average RBE calculated on all the RBE values was 3.1 ± 3.7 (standard deviation).

Other data meta-analyses have derived RBE factors greater than 1 for tritium. Based on a review of tritium effects on fish and mammals, Environment Canada (2000) recommended the adoption of an RBE factor of 3 for tritium. UNSCEAR (1996) recommended an RBE factor for β radiation of 2 for low energy β particles (<10 keV), and 1 for energies greater than 10 keV. In the framework of European programs FASSET and ERICA (FASSET 2003, 2004; ERICA 2006), a similar recommendation was made, with a RBE value of 3 for β radiation particles of low energy.

Ionizations caused by tritium are at a relatively high density and are thus likely to lead to significant damage. Additionally, there are two further theoretical explanations as to why an RBE greater than 1 might be expected for tritium: possible effects of transmutations to helium (e.g., leading to excess mutations) and accumulation of tritium in the hydration shell of DNA, named “buried tritium” (HPA 2007). In such a fraction, tritium is located in bridge positions wherein the exchange rates are reduced from microseconds to days, months, or even years, thus enhancing the probability of effects occurring.

However, our review has highlighted some very low EDR_{10} values obtained for tritium effects in invertebrates, sometimes even lower than the external γ -irradiation

background dose rate. It is difficult to understand how effects could be significant at such low levels. All species have had to evolve in a background of natural cosmic and terrestrial radiation. As a result, all organisms have efficient mechanisms to repair cells damaged from irradiation, or that promote cell apoptosis such that the damage is not propagated. Similar mechanisms exist and are necessary to repair the naturally occurring oxidative damage produced as a by-product of normal metabolism. Such low EDR_{10} values suggest that additional work is required to substantiate the original research. One of the possible explanations may be that dose rates were underestimated or were inadequate. Indeed, the dose rate either calculated or given by original authors assumes a homogenous distribution of tritium in the organisms they test. However, if dose distribution is relatively homogenous for tritiated water, it is heterogenous when tritium is incorporated into proteins and DNA. Therefore, the reliability of the concept of average dose to organisms must be questioned, and addressed.

5.1.3 Vertebrate vs. Invertebrate Sensitivity

Results obtained with HTO suggest that invertebrates are more sensitive than vertebrates, although this conclusion is contrary to the established paradigm (based on LD_{50}) that invertebrates are more resistant to radiation types. It is possible that problems exist with dosimetry, as described in the previous section. It is also possible that the higher radiosensitivity of invertebrates may be linked to the way they develop. Indeed, invertebrates generally undergo determinate development or mosaic development, in which cell lineage is determined primarily by the genome and cytoplasm of each individual cell. As a consequence, if a cell dies during development, then none of the tissues that would have formed from the progeny of that cell can develop. This also takes place in nematodes like *C. elegans*, which show almost no ability to compensate for the exposure-induced deletion of individual cells during development. In contrast, vertebrates undergo indeterminate or regulated development, and cell lineage is strongly determined by the interaction of the genome with epigenetic (nongenetic) factors extrinsic to the individual cell, such as molecules released by neighboring cells. Epigenetic factors interact with the cell's genome to determine its fate, through a process known as induction. Because any cell's fate is the consequence of induction, removal of a few cells will not adversely affect development since other cells will simply be induced to take over for the missing ones.

5.2 Biological Endpoint Sensitivity

There are some fundamentals that emerge from this review in terms of biological effects. Foremost is that there is not one universal endpoint to study. Additionally, regardless of the endpoint chosen, a considerable range in sensitivities occurs.

Different sensitivities appear depending on the tissue considered, the sampling time, or the development time. For example, there are several cases in which responses decrease with dose rate (e.g., SCE, malformations—Hagger et al. 2005), but these cases can be explained by compensation mechanisms (repair or apoptosis).

Hence, it is highly important to combine several responses at different periods of the cell or life cycle to be able to understand the health consequences of observed effects.

Studies performed on development highlight the importance of considering a long exposure time, and to include a parental exposure, rather than forming conclusions based solely on an embryonic exposure (e.g., Harrison and Anderson 1988). A wide diversity of reproductive effects were observed, such as ovotestes (Egami and Hyodo-Taguchi 1969), changes in sex ratios and courting behavior (Erickson 1971), and decrease in testes mass (Knowles 1999). Interesting results obtained on guppies (Erickson 1971) indicate that ionizing radiation may affect the magnitude or timing of hormone production by the pituitary, and directly and indirectly, testosterone production by the testes.

5.3 Biomonitoring and Risk Assessment

In this chapter, we have seen how reproductive toxicity and genotoxicity can be used to assess the impact of radionuclides under laboratory conditions on aquatic organisms. It is remarkable that endpoints measured to address reproductive toxicity were used much more frequently in the laboratory than in the field, and vice versa for genotoxicity endpoints (Adam 2007; Geras'kin et al. 2008). This may result from the relative difficulty in measuring endpoints such as brood size and embryo/larvae viability in the field (except for ovoviviparous species such as the mosquitofish).

One of the objectives of this review was to focus on the genotoxicity of radionuclides and the downstream relevance to reproduction, development, and survival of early-life stages. However, these two types of endpoints were very rarely measured together, except by Theodorakis et al. (1997), who demonstrated a clear relationship between DBSs and reproductive effects.

Genotoxicity endpoints present several advantages: (1) sampling is nondestructive for blood or sperm; (2) they are easier and more rapidly assessed compared to histological analyses of gonads, for example; and (3) they are sensitive to radionuclides (e.g., significant effects on gametes at a dose of 1 mGy/day of γ irradiation). Moreover, although little is known about the transgenerational effects of mutations in germ cells, they are probably the most meaningful among the other subcellular endpoints. However, their significance to organisms and populations is less trivial than is a reproductive endpoint. It is difficult to evaluate all the significance of endpoint measurements relative to the health of an entire ecosystem. One of the most popular tools to assess the impact of radionuclides on the environment is to perform an ecological risk assessment, by using screening calculations, in which radionuclide

levels in various environmental compartments are compared against benchmarks. In contrast, biomonitoring programs for sites with radionuclide contamination consists of sampling environmental compartments (e.g., water, sediments, and soils) and tissue residues of various species. Analyses of such samples only provide a snapshot of current internal dose rates and do not provide information on the external dose rate. Moreover, they may reveal only a part of the real exposure of an organism. This is because it is expensive to systematically measure all contaminants (not only γ - but also alpha-emitters, as well as chemical trace metals, organic pollutants, etc.). Use of a direct biomarker of genetic damage in species of concern could provide a meaningful indicator of biological damage. Furthermore, if this biological or genetic damage has potential reproductive effects, it could be an ecologically relevant assessment endpoint (e.g., Ulsh et al. 2003), and could be used as a prognostic biomarker, as is done in human health. Such a system could provide an early-warning of ecosystem injury (Moore et al. 2004), if used together with other pathological cellular changes (e.g., histopathology of gonads, lysosomal stability, immunotoxicity) in a holistic approach that properly integrated prognostic biomarkers and generic simulation models. However, more effort is needed to apply these techniques directly in field testing in which the variability of responses to natural environmental stressors must also be considered.

5.4 Research Directions

In the domain of evaluating the ecotoxic risks faced by aquatic organisms from exposure to radioactive pollutants, there are four relatively unexplored topics that concern different biological orders. We address these below and summarize our views on the needs for additional research for each of them:

1. *DNA double strand break characterization and repair.* Knowledge on the formation of DNA DBSs that are induced by ionizing radiation and their repair has progressed considerably over the last 10 years. DSB repair by the nonhomologous end-joining (NHEJ) pathway in vertebrates requires at least four gene products: Ku80, Ku70, DNA ligase IV, and the DNA-dependent protein kinase (DNApK) catalytic subunit (Weterings and Chen 2008). This repair pathway is conserved in mammals. Putative orthologs for each of these were identified in the zebrafish EST database and were studied (Bladen et al. 2005, 2007). The presence of these genes, together with the functional characterization of the Ku70 and Ku80 genes in the zebrafish, suggest that an intact functional NHEJ probably operates in the zebrafish and also in other fish species.

Despite the existence of several studies on DNA damage and repair in different fish species (e.g., Kosmehl et al. 2008; Sandrini et al. 2009; Cambier et al. 2010), very few data are available on the most deleterious effects, viz., DNA DBSs. Identification of impaired DNA DBS repair pathways and kinetics can be determined by using the number of nuclear foci formed by the phosphorylation

of the variant histone H2AX (pH2AX), easily quantifiable by employing immunofluorescence on cells. Such a technique has been successfully tested and was presented as a powerful predictive assay of radiosensitivity for mammalian cells (Joubert and Foray 2006). Indeed, some data showed that anti-pH2AX immunofluorescence does not necessarily predict the whole range of human radiosensitivity and that detection of other DNA repair proteins such as DNAPK are necessary to understand and explain radiation-induced NHEJ defects at the cellular level (Joubert and Foray 2006).

These methods and their results can be relevant to fish species and will be helpful to establish the dose-dependent correlation between DNA damage accumulation and abnormalities in embryo development (Pereira et al. 2011). In time, DNA repair proteins could be validated as predictive biomarkers of developmental or reproductive effects. Such biomarkers could have important implications for environmental protection from ionizing radiation exposure, because they may be more sensitive than other macroscopic endpoints (Anderson and Wild 1994).

2. *Transgenerational effects.* The transmission of genetic damage to offspring is of primary concern in the human health arena. However, there has been little work undertaken to assess the potential risk from germ cell mutagens in aquatic organisms, although this is one of the means of extrapolating effects from sub-cellular levels to populations. As described in Sect. 4 of this review, there have been some studies performed that used very specific techniques to understand the transgenerational effects of different types of radiation. In one study performed over five generations, chronic exposure of medaka to external γ irradiation resulted in significant differences in reproductive effects that were attributed to the accumulation of total dose (over 3 Gy) (Hinton et al. 2011). Shimada and Shima (2004) have developed a non-mammalian system, using the medaka fish, to analyze mutants as mosaics of orange and white pigment cells. Tsyusko et al. (2007) have also developed a system in the same fish species to study mutation rates in microsatellites of parents and offspring. If these techniques can be used to evaluate the potential hazard of substances in the laboratory, their application to a wide range of natural species may be very difficult since their genetic characteristics are generally poorly known (e.g., microsatellites, cell turn-over rate, or sensitivity). Other methods, such as the assessment of genetic change via allozyme survey and molecular techniques such as RAPD may be used for that purpose (Jha 2004).
3. *Reproductive behavior and endocrine function.* Despite the importance of behavior in the reproduction of fish, only one study has been performed on this topic in the field of radionuclides (Erickson 1971), whereas this endpoint has been widely used for endocrine disruptors (e.g., Baatrup 2009). In fish exposed to γ -rays, courtship behavior decreased with increasing dose rates (Erickson 1971), and in fish exposed to HTO, there are strong presumptions that male courtship behavior was altered, since the oviposition of nonexposed females was reduced in the presence of exposed males (Hyodo-Taguchi and Etoh 1986).

Furthermore, there are no studies of radionuclide effects on endocrine function, despite the fact that some results suggest reproduction is altered through effects

on testosterone. The developments that have taken place recently in the field of endocrine disruptors could be used to better assess both the direct or indirect radionuclide effects on endocrine function.

4. *Adaptation/acclimation.* Despite the existence of chronic exposure to radionuclides at levels eliciting toxicities in naïve-populations (e.g., Chernobyl or areas in Kazakhstan), organisms often thrive in highly contaminated environments, because they develop resistance to the toxic effects. It is important to distinguish between the two main possible forms of resistance. Acclimation occurs at the organism level, and is manifested through physiological or epigenetic mechanisms that ameliorate the toxic effects of contaminant exposure. Since such acclimation is not transmitted through generations, it should disappear in remediated environments. Adaptation occurs at the population level, with the genetic selection of the more resistant organisms. Using wildtype and radiosensitive strains of medaka exposed to 0.5 Gy of γ irradiation, proteomic changes indicated both immediate protection and longer term adaptation to subsequent radiation exposure (Smith et al. 2011). Although evolutionary changes are thought to require long time scales, adaptation can occur rapidly if the selection pressure is high. For radionuclide-contaminated sites, there is only one study showing adaptation in pond snails in Chernobyl (Golubev et al. 2005). Given that invertebrates have a shorter generation time, are generally exposed to higher dose rates and are less mobile than fish, the probability that they develop adaptation is higher than in fish (Jha 2004). However, different resistance strategies (adaptation vs. acclimation) were demonstrated in fish populations exposed to organic micropollutants (Wirgin and Waldman 2004), and could be studied in the context of radioactive contamination. It is therefore recommended that adaptation strategies should be studied in invertebrates and vertebrates, and, moreover, the recent Fukushima disaster could offer the opportunity to evaluate these mechanisms in the field.

6 Summary

Aquatic ecosystems are chronically exposed to natural radioactivity or to artificial radionuclides released by human activities (e.g., nuclear medicine and biology, nuclear industry, military applications). Should the nuclear industry expand in the future, radioactive environmental releases, under normal operating conditions or accidental ones, are expected to increase, which raises public concerns about possible consequences on the environment and human health.

Radionuclide exposures may drive macromolecule alterations, and among macromolecules DNA is the major target for ionizing radiations. DNA damage, if not correctly repaired, may induce mutations, teratogenesis, and reproductive effects. As such, damage at the molecular level may have consequences at the population level. In this review, we present an overview of the literature dealing with the effects of radionuclides on DNA, development, and reproduction of aquatic organisms.

The review focuses on the main radionuclides that are released by nuclear power plants under normal operating conditions, γ emitters and tritium. Additionally, we fitted nonlinear curves to the dose–response data provided in the reviewed publications and manuscripts, and thus obtained endpoints commonly associated with ecotoxicological studies, such as the EDR_{10} . These were then used as a common metric for comparing the values and data published in the literature.

The effects of tritium on aquatic organisms were reviewed for dose rates that ranged from 29 nGy/day to 29 Gy/day. Although beta emission from tritium decay presents a rather special risk of damage to DNA, genotoxicity-induced by tritium has been scarcely studied. Most of the effects studied have related to reproduction and development. Species sensitivity and the form of tritium present are important factors that drive the ecotoxicity of tritium. We have concluded from this review that invertebrates are more sensitive to the effects of tritium than are vertebrates. Because several calculated EDR_{10} values are ten times lower than background levels of γ irradiation the results of some studies either markedly call into question the adequacy of the benchmark value of 0.24 mGy/day for aquatic ecosystems that was recommended by Garnier-Laplace et al. (2006), or the dose rate estimates made in the original research, from which our EDR_{10} values were derived, were underestimated, or were inadequate.

For γ irradiation, the effects of several different dose rates on aquatic organisms were reviewed, and these ranged from 1 mGy/day to 18 Gy/day. DNA damage from exposure to γ irradiation was studied more often than for tritium, but the major part of the literature addressed effects on reproduction and development. These data sets support the benchmark value of 0.24 mGy/day, which is recommended to protect aquatic ecosystems.

RBEs, that describe the relative effectiveness of different radiation types to produce the same biological effect, were calculated using the available datasets. These RBE values ranged from 0.06 to 14.9, depending on the biological effect studied, and they had a mean of 3.1 ± 3.7 (standard deviation). This value is similar to the RBE factors of 2–3 recommended by international organizations responsible for providing guidance on radiation safety.

Many knowledge gaps remain relative to the biological effects produced from exposure to tritium and γ emitters. Among these are:

- Dose calculations: this review highlights several EDR_{10} values that are below the normal range of background radiation. One explanation for this result is that dose rates were underestimated from uncertainties linked to the heterogenous distribution of tritium in cells. Therefore, the reliability of the concept of average dose to organisms must be addressed.
- Mechanisms of DNA DBS repair: very few studies address the most deleterious form of DNA damage, which are DNA DBSs. Future studies should focus on identifying impaired DNA DBS repair pathways and kinetics, in combination with developmental and reproductive effects.
- The transmission of genetic damage to offspring, which is of primary concern in the human health arena. However, there has been little work undertaken to assess

- the potential risk from germ cell mutagens in aquatic organisms, although this is one of the means of extrapolating effects from subcellular levels to populations.
- Reproductive behavior that is linked to alterations of endocrine function. Despite the importance of reproduction for population dynamics, many key endpoints were scarcely addressed within this topic. Hence, there is, to our knowledge, only one study of courtship behavior in fish exposed to γ rays, while no studies of radionuclide effects on fish endocrine function exist. Recent technical advances in the field of endocrine disruptors can be used to assess the direct or indirect effects of radionuclides on endocrine function.
 - Identifying whether resistance to radiation effects in the field result from adaptation or acclimation mechanisms. Organisms may develop resistance to the toxic effects of high concentrations of radionuclides. Adaptation occurs at the population level by genetic selection for more resistant organisms. To date, very few field studies exist in which adaptation has been addressed, despite the fact that it represents an unknown influence on observed biological responses.

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