SAFE SEAFOOD GUIDE FOR POLICYMAKERS:
EMERGING CHEMICAL CONTAMINANTS IN SEAFOOD

JANUARY 2017
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INTRODUCTION

Seafood is widely recognised as a high quality and healthy food source. European Union (EU) legislation ensures that food placed on the market in Europe is safe to eat and does not contain contaminants at levels that could threaten human health. However, new seafood health risks from contaminants have emerged, for which limited information is available and for which authorities have not yet set maximum limits.

In response to these emerging risks, research has been committed to understanding these risks in relation to seafood consumption. ECsafeSEAFOOD, a European Union-funded research project dedicated to assessing potential risks of seafood consumption and ways of minimising them, was one such initiative. ECsafeSEAFOOD developed this guide for policymakers based on scientific evidence from the project to:

- effectively communicate the risks of chemical contaminants of emerging concern in seafood and their impact on public health across the EU;
- provide information that may help inform the development of new or improved rules to manage the risk of emerging hazards in seafood;
- provide information that may eventually inform setting permissible levels of emerging chemical contaminants in seafood products, while accounting for the effects of culinary treatment and processing on contaminant levels;
- support the implementation of mitigation strategies for any negative impact contaminants may cause.

This guide informs policymakers and food safety authorities of the latest seafood safety research results. We hope this information will contribute to European science-based food safety regulation, specifically in relation to newly emerging chemical contaminants which have not yet been regulated.

Throughout its work, the ECsafeSEAFOOD project has addressed the Marine Strategy Framework Directive (MSFD) - European legislation that aims to achieve Good Environmental Status (GES) of EU marine waters by 2020. The Directive defines Good Environmental Status as:

"The environmental status of marine waters where these provide ecologically diverse and dynamic oceans and seas which are clean, healthy and productive."

To help Member States interpret what GES means in practice, the Directive sets out 11 qualitative descriptors that describe what the environment will look like when GES has been achieved. In particular, the ECsafeSEAFOOD project has addressed Descriptor 9 (contaminants in seafood are below safe levels) through assessments of the levels of emerging chemical contaminants in seafood, and Descriptor 10 (marine litter does not cause harm) through the assessment of microplastics in seafood.

Seafood Health Benefits

According to recent results from the European Food Safety Authority (EFSA, 2014), seafood has many health benefits:

- **Seafood is an excellent source of energy and protein, containing all essential amino acids humans need;**
- **Seafood contributes to the intake of essential nutrients, such as iodine, selenium, calcium, and vitamins A and D, which have well established health benefits;**
- **Seafood is a valuable source of minerals and trace elements such as zinc, phosphorus, iron and copper, which are required by the body for a variety of functions;**
Seafood provides n-3 long-chain polyunsaturated fatty acids, which in contrast to other nutrients are obtained mainly from seafood. These include omega-3 fatty acids required for normal growth and to support immunity, and which can improve cardiovascular and brain health;

- In the general adult population, consumption of seafood, particularly fatty fish, lowers the risk of mortality from coronary heart disease;
- Consumption of about one to two servings of seafood per week, and up to three to four servings of seafood per week during pregnancy, has been associated with better functional outcomes of neurodevelopment in children compared to no consumption of seafood during pregnancy. However, during pregnancy, consumption of seafood species with a high mercury content (e.g. tuna, swordfish) should be limited and replaced with species low in methylmercury (e.g. sardines, anchovies, mackerel, trout);
- Seafood is a component of dietary patterns associated with good health.

Most European food-based dietary guidelines recommend two servings of seafood per week. This ensures the provision of key nutrients, especially omega-3, but also vitamin D, iodine and selenium.

**Seafood Health Risks**

Although the consumption of seafood (following official recommendations) has many health benefits for humans, like other food types, it can also pose a potential health risk. Seafood health risks can arise from environmental contamination (e.g. natural toxins, metals, plus pollutants) and can be introduced directly by human activities, including the mishandling of food during preparation.

In Europe, all seafood intended for human consumption must comply with general health requirements ensuring that all seafood for sale is of good standard. Legislation has restricted contaminants that may have otherwise ended up in seafood from various stages in the production process or from contamination in the marine environment. Each EU country carries out a range of seafood monitoring practices to ensure that all seafood products available in the market comply with the highest food safety standards.

Research forms the backbone of the EU’s legislation on contaminants and EU measures are regularly reassessed through risk assessments by EFSA in light of the most recent scientific knowledge. The European Commission co-funds various research projects on this issue, including the ECsafeSEAFOOD project. ECsafeSEAFOOD has focused on the issue of seafood safety in relation to what are known as “priority contaminants”, present in seafood as a result of environmental contamination. Priority contaminants are contaminants for which no maximum limits have yet been set by authorities and that are not yet controlled by monitoring programmes. Priority contaminants investigated by ECsafeSEAFOOD include those originating from pharmaceuticals, personal care products and brominated flame retardants, as well as those originating from harmful algal blooms (marine biotoxins) and those associated with marine litter.

This ECsafeSEAFOOD guide informs policymakers about the latest research results in relation to newly emerging chemical contaminants that can be found in seafood. Through this guide, the ECsafeSEAFOOD project hopes to inform evidence-based policymaking related to this topic.
ECsafeSEAFOOD research results show that seafood consumption varies widely between the different EU countries, mainly because of country-related traditions, diverse eating habits and availability of seafood. In general, people living less than 20 kilometres from the coast consume more seafood than people living further inland. Seafood is bought most frequently at supermarkets, followed by fishmongers. When comparing the different types of seafood, consumers show the most positive attitude towards finfish and the least positive attitude towards seaweeds, followed by farmed fish. Finfish are also perceived by consumers as the healthiest seafood, of high quality, nutritious, and safer than meat. While consumers may be less likely to buy farmed fish as they have a less positive attitude towards it, farmed fish is perceived as the most affordable seafood. Respondents in Italy, Spain, and Portugal had a positive attitude towards crustaceans or shellfish, which differs from the respondents in Ireland and Belgium who had a more neutral attitude towards crustaceans.

Detailed information on these studies can be found in the scientific paper by Jacobs et al. (2015) (see References, page 17 for more).

Consumer Perception of Marine Environmental Contamination

ECsafeSEAFOOD research results show that consumers are concerned about marine environmental problems and attribute the contaminants in the marine environment especially to industrial pollution, followed by human domestic waste. People from southern European countries report a stronger concern about the marine environment compared with citizens from northern and western European countries. Additionally, people who live closer to coastal areas also show stronger concern about the consequences of marine environmental problems. In general, people are more concerned about the consequences for the global ecosystem, such as sea life, than about direct consequences to themselves and others.

Consumers do not have a strong belief that they are able to help solve marine environmental problems at an individual level. However, when they are challenged to take a concrete action, such as eating more sustainable seafood, their confidence in the potential impact of individuals on the environment increases. It is clear that for environmental policy-makers and organisations aimed at stimulating pro-environment behaviour in consumers, it is not enough to raise awareness about marine environmental problems and the possible impacts of consumer behaviour. Their message for consumers must also contain a concrete or tangible action.

More information can be found in the following scientific paper: Jacobs et al. (2015) (see References, page 17 for more).

Policy Recommendations

- Implement mitigation measures for emerging chemical contaminants in seafood;
- Implement campaigns to warn consumers of the need to diversify food consumption, reduce consumption of predatory species and increase consumption of low-trophic level species (i.e. species that are low down in the food-chain);
- Funding is needed to better understand the accumulation of seafood contaminants and the toxicity of contaminants in humans.
Emerging Seafood Safety Risks
Emerging Chemical Contaminants

Problems related to chemical contamination of the environment are nearly all caused by humans. The dumping of hundreds of millions of tonnes of material from industrial processing, sludge from sewage treatment plants, agricultural drainage and raw untreated sewage into the ocean contributes to the contamination of coastal marine and freshwater environments. Various chemicals find their way from the aquatic environment into fish and other aquatic organisms.

ECsafeSEAFOOD has carried out research on emerging chemical contaminants, including:

- Pharmaceuticals (PhACs) (e.g. the anti-inflammatory drug diclofenac, the sedative diazepam, the antibiotic azithromycin);
- Personal care products (e.g. galaxolide (HHCB) - a synthetic musk/fragrance used in cosmetics; 2-ethyl-hexyl-4-trimethoxycinnamate (EHMC) - a UV filter found in sunscreen);
- Toxic elements and their species (e.g. inorganic arsenic, organic mercury, cadmium) – elements that occur naturally but become concentrated as a result of human activities such as mining and some industrial processes;
- Polycyclic aromatic hydrocarbons (PAHs) – a group of chemicals that are released from burning coal, oil, gasoline, rubbish, tobacco, wood, or other organic substances;
- Brominated flame retardants (BFRs) – man-made chemicals used commonly in plastics, textiles and electrical/electronic equipment (e.g. polybrominated diphenyl ether (PBDE99));
- Perfluorinated compounds (PFCs) – a large group of manufactured compounds that are widely used to make everyday products more resistant to stains, grease and water. They are also used in the aerospace, automotive, building and construction, and electronics industries;
- Plasticizers (e.g. bisphenol A (BPA), phthalates, polychlorinated biphenyls (PCBs)) – substances that are added to a material to increase their flexibility, usually a plastic.

What is Being Done

Limits have only been set for a certain number of contaminants like some toxic metals (total mercury (Hg), cadmium (Cd), lead (Pb)), dioxins and PCBs and some toxins. For these contaminants there are specific monitoring programmes in each EU country (screened by the European Commission).

The emerging contaminants are not regulated and therefore not monitored yet.

Policy Recommendations

The ECsafeSEAFOOD project makes the following recommendations to policymakers based on the project results in relation to emerging chemical contaminants:

- Monitor specific contaminants like emerging toxins PBDE, MeHg and iAs on a regular basis;
- Make information on environmental hazards and chemicals as emerging seafood safety risks transparent and readily available to stakeholders;
- Ensure validation and recognition of novel fast screening tools for emerging chemical contaminants (including those developed in ECsafeSEAFOOD, other EU-funded projects and other organisations), to complement conventional contaminant screening methodologies;
- Consider regulations on emerging chemical contaminants methylmercury (MeHg) and pentabromodiphenyl 99 (PBDE99) where the exposure to consumers is at a concerning level;
- Support further research that evaluates the toxic effect of both arsenite and arsenate, as separate inorganic arsenic species, because initial research has highlighted there is a significant difference in toxic effects;
- Support more research (funding) in the field of seafood safety to identify and establish limits for emerging chemical contaminant levels in seafood. Once limits are established, seafood should be monitored where there is risk of contamination to ensure these limits aren’t exceeded and that seafood is safe for consumption.
Microplastics

More than 80% of the waste that ends up in our world’s oceans is generated on land. One of the main contributors to this waste is plastic. Plastic does not biodegrade easily and stays in our environment for a very long time, causing a serious problem. Nowadays many plastic recycling initiatives reduce the amount of plastic that ends up in the environment, ultimately keeping it out of the ocean. However, there is a certain type of plastic whose damaging effects to sealife are not yet fully understood: microplastics.

Microplastics:
• are small plastic particles (generally smaller than 5 mm);
• come from a variety of sources including personal care products like toothpaste, cleaning agents and industrial processes; and
• have been found in seafood (such as fish, shrimp, and some shellfish), in non-seafood products (e.g. honey, beer and table salt) and even in the air we breathe.

There is concern that these microplastics can accumulate up the marine food chain, and the risk is that humans may eventually ingest these microplastics when they eat (sea)food contaminated with microplastics. At the moment, researchers do not fully understand the fate and toxicity of microplastics in humans, whatever way they have been ingested (EFSA, 2016). A recent opinion of the Dutch Health Council came to a similar conclusion, meaning that it is difficult to make direct recommendations at this stage (Gezondheidsraad, 2016).

The latest research results suggest that the quantity of microplastics in the edible portion of fish is likely to be negligible for consumer exposure. This is because if a fish ingests microplastics, they will mainly be contained in the digestive tract, which is normally discarded before consumption. However, bivalves, such as mussels, oysters and crab, are filter feeders and can therefore accumulate microplastics - and, as opposed to fish, their digestive tract is eaten. Nevertheless, research suggests that microplastics may not be as harmful as previously thought.

At present, there is no knowledge on the fate of microplastics during the processing of seafood.

What is Being Done

There is no current EU-wide legislation specific to microplastics as contaminants in food, including seafood. However, there is a broad range of EU policies and legislation with regard to marine litter, covering sources and impacts.

The use of microbeads in cosmetics has already been phased out by many cosmetic companies, and there are ongoing developments to ban the production and sales of personal care products containing microbeads completely.

Policy Recommendations

• Support further research (funding) to further investigate the fate of microplastics during the processing of seafood and whether there are any adverse health effects from microplastics (and nanoplastics) in seafood;
• Develop and implement a waste management plan to reduce plastic waste, and a recycling campaign to avoid litter on beaches and inland rivers;
• Recommend, encourage or incentivise the plastics industry and scientific research to produce ‘environment-friendly degradable plastics’, i.e. plastics that can be completely degraded in the occurring environmental conditions (improving on the current biodegradable plastics, which are often just partially degraded) or alternatives to plastic;
• Develop campaigns and communication strategies to increase consumer awareness about microplastics (e.g. how to avoid consuming them and help prevent microplastic pollution).
Biotoxins

The ECsafeSEAFOOD project also carried out research on marine biotoxins (e.g. azaspiracids (AZA), okadaic acid (OA), ciguatoxins (CTXs), tetrodotoxin (TTX)). Marine biotoxins are toxic substances produced by marine microorganisms, such as microalgae, which may cause harmful algal blooms (HABs). In recent years, Europe has experienced HAB events that have threatened public health and caused enormous economic losses to fisheries and tourism. These HAB events have been increasing in size and frequency globally. Marine biotoxins can occasionally contaminate some species of shellfish and, in some areas, fish and other organisms. As shellfish feed by straining suspended matter and food particles from water, they can accumulate marine toxins from the harmful microalgae present in the environment. The consumption of seafood contaminated with biotoxins can be harmful to human health, with the risk and severity of poisoning being very serious.

Public health authorities worldwide have established regulations and risk management plans to deal with these marine biotoxins. EU-wide legislation limits levels of marine biotoxins in seafood that are to be placed on the market. Seafood produced or caught in the EU, and in non-EU countries exporting their seafood into the EU, is regularly monitored for known biotoxins. All shellfish production areas in the EU are also regularly monitored. So, regardless of origin and species, seafood commercialised through the appropriate channels in the EU should be safe for consumption. If contamination risks are identified, local authorities will implement management actions such as closing shellfish sites or banning the sale of certain species until tests prove the risk has passed, in order to protect the consumer.

A relatively new emerging biotoxin present in some species of fish in Europe is ciguatoxin (CTX), causing ciguatera in humans, resulting in gastrointestinal, cardiovascular and neurological problems. During the last decade ciguatera has been reported in the Canary Islands and Madeira, two areas that were previously not affected. In these areas several ciguatera poisonings have been reported after consumption of fish, mainly amberjack (Seriola sp.) since 2008.

Pufferfish (Tetrodontids) constitute a food safety concern since they may contain dangerous amounts of the neurotoxins known as tetrodotoxins (TTXs). EU legislation is clear, and pufferfish cannot be placed on the market in Europe at all. A highly toxic species of pufferfish (Lagocephalus sceleratus) has recently invaded the Mediterranean coming from the Suez Canal, and is spreading across the Mediterranean.

Between 2014-2015, TTXs have been found in bivalve mollusc shellfish grown in the south of England, along the Greek coast, and in the Netherlands (Vlamis et al 2015, Turner et al 2014, RASFF Consumers Portal 2016).

Cyclic imines are newly emerging neurotoxins that can be present in seafood. They have been detected in several species of fresh shellfish and processed seafood. Information on their toxicity and distribution is needed in order to understand their real risk. Since the exact risks associated with these toxins have not yet been characterised, research efforts should be focused on assessing their occurrence and toxicity.

What is Being Done

All EU countries that have shellfish harvesting areas run biotoxin monitoring programmes to ensure that seafood from these areas complies with EU biotoxin regulations (e.g. Regulation EC No 853/2004 governs the total amount of marine biotoxins that may be present in shellfish). Water quality testing is carried out by national reference laboratories, as well as by shellfish farmers themselves, as part of legislative requirements. In addition, monitoring for biotoxins is conducted on all other types of seafood sold in the EU.

However, in relation to ciguatoxins, currently there are no regulatory limits for CTX-group toxins in the EU, even though regulation states that checks are to take place to ensure that fishery products containing biotoxins such as ciguatoxin are not placed on the market. Up until now, none of the current methods of analysis to determine CTX-group toxins has been formally validated in Europe. To allow validated method development, certified reference standards and reference materials need to be provided.
The ECsafeSEAFOOD project has successfully developed and applied the use of a cell-based assay and an analytical method based on liquid chromatography coupled to mass spectrometry (LC-MS/MS) for the identification and quantification of CTXs. This approach has been recommended for consideration by Codex Alimentarius or “Food Code” (established by FAO and the World Health Organization in 1963 to develop harmonised international food standards) and is also already successfully implemented in several other countries, including the USA (by the Food and Drug Administration). ECsafeSEAFOOD therefore recommends formal validation of the LC-MS/MS approach (besides the neuroblastoma (Neuro-2a) cell-based assay) to identify and quantify CTX-group toxins in Europe. Such validation where these issues are partially covered, is being performed as part of the EuroCigua project—a collaboration from EFSA and 14 other partners on the characterisation of ciguatera food poisoning in Europe.

Regional public health authorities in the Canary Islands and Madeira have set up management procedures through local regulations in order to limit the consumption of fish that have a high risk of being contaminated with ciguatera, and a monitoring programme for ciguatoxins has been established in the Canary Islands.

Regarding tetrodotoxins (TTX) and cyclic imines, these toxins are the subject of several research projects to assess and understand their risk. Pufferfish, being carriers of TTX, have been banned for consumption in the EU. However, given that pufferfish is sometimes offered at private events, the risk may not be completely disregarded and should be assessed. TTX poisoning episodes have also been reported following the consumption of mislabelled fish products and, additionally, pufferfish may be consumed by accident due to their similarity to other non-poisonous fish. Currently, there is no official method of analysis for TTX in Europe, but the most widely employed methods are the mouse bioassay and LC-MS/MS. Although proven to be useful techniques, the need for skilled personnel, expensive equipment and standards of TTX and TTX analogues for LC-MS/MS, added to the lack of specificity of the frequently used mouse bioassay in discriminating between other coexisting paralytic shellfish poisoning toxins, have hampered their performance. Due to these limiting factors, and given the increasing occurrence of TTX in Europe, the development of specific, rapid and cost-effective methods as support tools in monitoring programmes is required to ensure human safety. The ECsafeSEAFOOD project has developed a specific, selective, rapid and easy-to-use assay based on antibodies for the detection of TTXs (mELISA), which is a promising tool for integration into monitoring programmes.

ECsafeSEAFOOD recommends more research into developing this fast screening method further. Recently, the European Commission has requested the European Food Safety Authority to provide a scientific opinion on the evaluation of the toxicity of TTX and TTX-analogues in bivalve molluscs and marine gastropods (EFSA 2016 – mandate: M-2016-0134). Cyclic imines can also be successfully identified by LC-MS/MS. The ECsafeSEAFOOD project recommends the implementation of multi-toxin identification methods for lipophilic toxins, which are already in place for the evaluation of regulated toxins, to gather data on the presence of cyclic imines. Should these toxins prove to be a hazard for consumers, fast screening methods currently being developed (e.g. by NVI in Norway) should be adapted for screening of these toxins.
Policy Recommendations

• In relation to ciguatoxins (CTXs), ECsafeSEAFOOD research results suggest that current guidance levels established by the US Food and Drug Administration (FDA) and European Food Safety Authority are appropriate, based on evaluation of CTXs in fish responsible for human poisoning;
• ECsafeSEAFOOD research has also demonstrated, as expected, that microalgae of the genus Gambierdiscus produce CTXs. It is therefore recommended that relevant governing bodies closely monitor CTXs in both the environment of current areas where exposure has taken place (Madeira and the Canary Islands), as well as in seafood from these areas. In particular, the temporal and spatial distribution of Gambierdiscus species populations in these areas should be monitored;
• Based on the above, it is recommended to support further research (funding) to be able to perform risk assessment studies and eventually establish appropriate regulations in the regions where CTXs are (becoming) prevalent, especially in areas where Gambierdiscus species have been found (e.g. Balearic islands, Greece, Cyprus);
• In relation to tetrodotoxin (TTX), based on incidents that have occurred and the recent invasion of the highly toxic species Lagocephalus sceleratus in the Mediterranean Sea, it is highly recommended to support further research (funding) in this area, and to make the public aware of the risks in the relevant areas;
• In relation to cyclic imines, it is recommended to support further research (funding), targeted towards assessing their occurrence and toxicity.

New Online Database of Emerging Chemical Contaminants in Seafood

The ECsafeSEAFOOD contaminants database is a unique database that collates data on emerging chemical contaminants in seafood species. The database focuses on the occurrence and effects of unregulated contaminants that give rise to concerns from an environmental and public health point of view. The contaminants in question include brominated flame retardants, endocrine disruptors, toxic element species, pharmaceuticals and personal care products, perfluorinated compounds, microplastics, and marine biotoxins among others.

The tool is useful for improving contaminant diagnostics and seafood risk assessment for food safety authorities and food diagnostic firms, as well as the seafood and aquaculture industries.

To request access to the ECsafeSEAFOOD Contaminants Database, please
Email: johanrobbens@ilvo.vlaanderen.be
Visit: www.ecsafeseafooddbase.eu

ECsafeSEAFOOD Contaminants Database - possible application by policy stakeholders:

The tool is useful for food safety authorities to improve seafood risk assessment. The data can also be used by policymakers to help inform policy and advisory guidelines, and by funding authorities to highlight the deficits in seafood contaminant research. The database contains information about the latest available data, both about presence and about effects of emerging chemical contaminants, and is therefore a good tool to provide policymakers with the most up-to-date information. The database also includes the (analytical) data that was generated during the project.
Effect of Cooking on Harmful Contaminants in Seafood

The ECsafeSEAFOOD project looked into the effect of cooking on the levels of several environmental contaminants in various seafood species. The table below gives you an understanding of what happens with the level of some contaminants in certain types of seafood when you cook them.

### Seafood Consumption: Effects of Cooking (Steaming)

Cooking affects environmental contaminant levels of metals, (methylmercury, inorganic arsenic), perfluorinated compounds (PFCs), endocrine disruptors (EDCs: chemicals which can interfere with hormone systems e.g. pharmaceuticals, pesticides, plasticisers and dioxin compounds), brominated flame retardants (BFRs), musks, polycyclic aromatic hydrocarbons (PAHs), UV filters and biotoxins in seafood. Tested species were ranked according to the number of contaminants showing increased levels after cooking by steaming, from the lowest (canned mackerel) to the highest (mussels). The levels of contaminants after cooking do not present a potential health risk for consumers.

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<th>METALS</th>
<th>PFCs</th>
<th>EDCs</th>
<th>BFRs</th>
<th>MUSKS</th>
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<td>Canned Mackerel</td>
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 '>' steaming increases the concentration of the specific contaminant level in particular seafood species, however these levels do not represent a potential health risk for consumers.

For a more detailed table outlining the effect of cooking on specific contaminants, see: http://ecsafeseafood.eu/cookingcontaminants
Policy Recommendations
Reassess current monitoring programmes to ensure inclusion of all elements possibly affecting seafood safety (e.g. including a seafood heating step).

Effect of Climate Change on Behaviour and Transfer of Relevant Priority Contaminants in Seafood

Today, anthropogenic activities have contributed to two great environmental concerns: first, the higher levels of chemical contamination and, second, the dramatic effects of climate change such as seawater warming and acidification. Both factors strongly affect marine ecosystems and their effects are expected to worsen in the future, threatening marine species’ welfare, survival and, ultimately, seafood consumers’ health. Yet, information is still rather limited in terms of the impacts of climate change on seafood safety and human health implications.

ECsafeSEAFOOD results indicate that climate change effects, may facilitate the bioaccumulation of chemical contaminants (like toxic metals, pharmaceuticals and personal care products, flame retardants and perfluorinated compounds) in seafood and, on the other hand, may also reduce seafood’s ability to purify them. Such alterations in conditions can lead to an increased human exposure to chemical contaminants through seafood consumption, which will likely raise concerns about consumers’ safety and public health.

For more information on the effects of climate change on contaminants in seafood see ECsafeSEAFOOD deliverable 6.4 on www.ecsafeseafood.eu or contact António Marques (amarques@ipma.pt).

Policy Recommendations
The expected increase in human health risks due to seafood contamination from the effects of climate change highlights the need to:

• allocate further research efforts and resources in this area, addressing various chemical contaminants and toxin-producing marine microorganisms responsible for HABs, as well as the expected impact of climate change (warming, acidification, reduced oxygen levels) on seafood safety;
• identify particularly sensitive areas and ecosystems in order to prevent risks for human health associated with seafood consumption;
• revise and adapt the current recommendations/regulations for the presence of chemical contaminants in seafood to make it more rigorous and include limits for emerging chemical contaminants, considering the expected climate change effects.
## Detecting Emerging Chemical Contaminants in Seafood

Several novel methods have been developed for the detection of pharmaceuticals and endocrine disrupting compounds in seafood within the ECsafeSEAFOOD project:

<table>
<thead>
<tr>
<th>Method</th>
<th>Type of sample</th>
<th>Family</th>
<th>Compounds</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>UHPLC-MS/MS to detect pharmaceuticals</td>
<td>Fish</td>
<td>β-blockers, psychiatric drugs, antiplatelet agent, analgesics/anti-inflammatory, diuretic, anthelmintic, and asthma drugs</td>
<td>atenolol, carazolol, metoprolol, nadolol, propranolol, sotalol, carvedilol, citalopram, diazepam, 10,11-epoxy-carbamazepine, 2-hydroxy-carbamazepine, lorazepam, sertraline, venlafaxine, clopidogrel, codeine, diclofenac, hydrochlorothiazide, levamisole, salbutamol</td>
<td>Huerta et al. 2013</td>
</tr>
<tr>
<td>UHPLC-MS/MS to detect pharmaceuticals</td>
<td>Bivalves</td>
<td>antibiotics, psychiatric drugs, analgesics, anti-inflammatory, tranquilizer, calcium channel blockers, diuretic, and prostatic hyperplasia</td>
<td>roxithromycin, metronidazole, dimetridazole, sulfamethoxazole, N-acetyl-sulfamethoxazole, azithromycin, erythromycin, ofloxacin, diclofenac, hidrocodone, oxytocin, hydrochlorothiazide, tamsulosin and some of their major metabolites</td>
<td>Alvarez-Muñoz et al. 2015</td>
</tr>
<tr>
<td>QuEChERS and LC-MS for antibiotics</td>
<td>Fish, clams and mussels</td>
<td>antibiotics</td>
<td>azithromycin, clarithromycin, roxithromycin, spiramycin, tilmicosin, tylosin, tetracycline, clindamycin, lincomycin, sulfadimethoxine, sulfamerazine, sulfamethoxazole, sulfadiazine, sulfapyridine, sulfisomidin, sulfisoxazole, N-acetylsulfadiazine, N-acetylsulfamerazine, N-acetylsulfamethazine, metronidazole, metronidazole-OH, trimethoprim, chloramphenicol</td>
<td>Serra-Compte et al. 2016</td>
</tr>
<tr>
<td>QuEChERS and UPLC-MS/MS</td>
<td>Fish</td>
<td>triazoles, stimulants, hormones, flame retardants, plasticizers, antibacterials and preservatives</td>
<td>1H-benzo-triazole, caffeine, progesterone, levonorgestrel, tolytriazole, tris(2-chloroethyl) phosphate, tris(2-butoxyethyl) phosphate, tris(2-chloroisopropyl) phosphate, estrone, 17β-estradiol, estriol, 17α-ethinylestradiol, estrone-3-sulfate, bisphenol A, triclosan, methylparaben, ethylparaben, propylparaben, benzylparaben</td>
<td>Jakimska et al. 2013</td>
</tr>
</tbody>
</table>

**Policy Recommendations**

ECsafeSEAFOOD researchers recommend the formal approval (or standardised use) of the above detection methods for pharmaceutical and endocrine disrupting compound monitoring programmes in Europe.
New Biosensors to Detect Priority Contaminants in Seafood

In recent years, biosensors have emerged as some of the leading relevant diagnostic tools for food safety monitoring due to their specificity, ease of mass fabrication, cost and field applicability. Biosensors complement standard analytical methods for environmental and food applications. In the case of certain marine biotoxins, biosensors might become a feasible alternative to animal experimentation in bioassays. These new tools are highly valuable since they are suitable for routine testing, first screening of samples and field analysis. They can be easily implemented in industrial processes providing relevant information for the quality and safety control systems of the seafood industry.

As part of the ECsafeSEAFOOD project, novel biosensors have been developed for different targets such as marine biotoxins (tetrodotoxins, azaspiracids), antibiotics (sulphonamides) and tetrabromobisphenol A. These new biosensors have been validated using real seafood samples and results compared with reference methods. This work has shown that these new tools are suitable for the determination of target analytes with the sensitivity required to determine the compliance of regulatory levels in a quick and easy way.

Policy Recommendations

Biosensors are suitable tools for the screening of contaminants in seafood samples within monitoring programmes and by the seafood industry, and can reduce the cost of analysis. The new biosensors developed within the framework of the ECsafeSEAFOOD project have been validated and are recommended for use to detect different targets such as marine biotoxins (tetrodotoxins, azaspiracids), antibiotics (sulphonamides) and the brominated flame retardant, tetrabromobisphenol A. In the case of positive results (presence of contamination), official reference methods of identification (generally a chromatographic technique) are necessary to confirm the results and to comply with legislative requirements.

New Reference Material for Emerging Biotoxins

In the EU there is a lack of certified reference material for several marine toxins, including some toxins that are regulated and most of those that are unregulated. The availability of reference material is crucial for the implementation of research and official controls. Policymakers can help overcome the shortage by supporting the availability of such material (through funding, incentives etc.). The ECsafeSEAFOOD project has gathered a collection of non-certified reference material for emerging marine biotoxins in different forms:

- A collection of cultures of toxin-producing microalgae;
- Extracts from toxic microalgae cultures;
- Toxic shellfish and fish samples (flesh, liver, other viscera) obtained from coastal areas;
- Purified standards.

Policy Recommendations

The EU and associated states should make more certified reference material for marine toxins available to improve and support the evaluation of toxins present in food and the environment. The availability of certified reference materials will contribute to improved detection and quantification of a higher number of toxin derivatives. To cover this lack of certified reference material, the EC should consider taking the following actions:

- Increase the number of established cultures of toxin producing microalgae;
- Promote and fund large-scale production of toxins from these cultures;
- Promote and fund the collection of toxin-containing seafood;
- Promote and fund the purification of toxins from these sources of toxins;
- Obtain certified reference material for marine toxins.
Phycoremediation involves the use of algae for the removal or biotransformation of contaminants from aquatic systems. This technology is environmentally friendly and less expensive than most other contaminant reduction methods. **ECsafeSEAFOOD** carried out phycoremediation research, using the seaweeds *Saccharina latissima* and *Laminaria digitata*, and discovered that seaweeds can contribute substantially to the uptake of organic and inorganic contaminants from seawater.

**ECsafeSEAFOOD** research results showed:

- A strong decrease (more than 85%) in venlafaxine (VEN) from seawater was observed during the first 12 hours of an experiment with the seaweed *S. latissima*;
- Uptake of VEN is faster in *S. latissima* than in *L. digitata* (24 hours instead of 120 hours);
- *L. digitata* proved to be the better candidate for removal of arsenic from seawater (reduction of more than 75% after 360 hours of the exposure);
- Neither *S. latissima* nor *L. digitata* was able to decrease concentrations of tetrabromobisphenol-A (TBBPA) and bisphenol-A (BPA) in seawater.

Seaweeds can also slightly reduce the bioaccumulation of some contaminants in mussels:

- Best results were obtained for VEN, inorganic arsenic (iAs) and diflubenzuron, with decreases of 25%, 84% and 70%, respectively;
- Levels of lindane, copper (Cu) and cadmium (Cd) did not decrease in mussels with the presence of *L. digitata*;
- The presence of contaminants in the medium did not affect the physiological condition of the seaweeds.

**Policy Recommendations**

Phycoremediation by macroalgae has the potential to be a good mitigation technology to reduce the contaminant levels in bivalves and/or in a water environment. This tool can possibly decrease the risk for human health and improve confidence in seafood consumption. Further research in this area is recommended.
New In Vitro Protocols for Ecotoxicity, Bioavailability and Bioaccessibility in Raw and Cooked Seafood

Levels of contaminants detected in seafood do not reflect the quantity that will be released from food during the digestive process. Different contaminants have different tendencies for becoming available for absorption at the intestinal epithelium, and entering into the blood stream, i.e. the bioaccessible fraction of a contaminant. They also can differ in terms of the degree and rate at which a substance is absorbed into a living system or made available at the site of physiological activity. This will depend on the molecular characteristics of the compound under study. In general, hydrophobic contaminants like polyfluorinated and polybrominated compounds have high rates of absorption of the intestine. Methylmercury has high absorption (50-70%, even 100% as identified in vivo), inorganic arsenic has intermediate absorption (10-20%) and cadmium has very low (below 2%) absorption.

Data on toxicity (the degree to which something is toxic to or damages living cells), bioavailability (the degree and rate at which a substance is absorbed into a living system or made available at the site of physiological activity) and bioaccessibility (the potential for a substance to interact with and be absorbed by an organism) of specific contaminants have been traditionally obtained from animal and human population studies. Due to ethical concerns about experimental practices involving higher animals e.g. rats, alternative experimental models have become increasingly popular.

Toxicity determination of highly hydrophobic substances is still challenging due to difficulties related to solubility and stability of such compounds in aqueous media that is used for most biological models (i.e. cells lines and aquatic animals). Synergistic and antagonistic effects on toxicity between contaminants in mixtures can occur and they have largely not been determined yet.

Policy Recommendations

- To assess and determine toxicity, bioavailability and bioaccessibility of contaminants in seafood, the stability and solubility of each substance tested should be evaluated during the whole exposure time to provide reliable results independent of the biological model used;
- The assessment of bioaccessibility is fundamental for risk-benefit analysis of nutrients and contaminants associated with food consumption, providing more accurate indications about health effects to consumers, refinements of food safety legislation (Maximum Permissible Concentrations; Tolerable Weekly Intakes; Recommended Daily Intakes) and guidelines for policymakers, thus minimising under- or over-estimations of risks/benefits, and providing more realistic information;
- In toxicity and bioavailability studies, laboratory in vitro analysis should be used over animal studies where possible due to ethical issues associated with using higher animals in research. Alternative models have been shown to provide relevant toxicity information but further validation efforts are necessary. Further efforts are needed to enable the evaluation of real contaminated samples with in vitro and alternative models.
ECsafeSEAFOOD GENERAL RECOMMENDATIONS AND CONCLUSIONS

The safety of seafood consumers requires the implementation of policies in accordance with the relevance of risks. ECsafeSEAFOOD has better characterised these risks in relation to emerging chemical contaminants, and provided evidence for non-regulated hazards. It is expected that ECsafeSEAFOOD’s scientific contributions will have an impact on the decisions that policymakers will make on emerging chemical contaminants in seafood.

Seafood is a unique, high quality, safe and nutritious food item. A diet rich in seafood contributes to a healthy life. The ECsafeSEAFOOD project found that despite the potential for emerging chemical contaminants to be found in raw and cooked seafood, the levels are low and represent no potential health risks for consumers following the recommendations of health authorities by eating two servings of seafood per week.

The ECsafeSEAFOOD project recommends:

• To diversify seafood consumption in order to ensure a balanced and healthy diet, prioritising species occupying lower trophic levels (e.g. mackerel, sardines, bivalve molluscs), species from sustainable and responsible fishing and farming practices, and species harvested in areas subjected to stringent environmental regulation mechanisms;
• To support monitoring strategies by supporting research to obtain more data on the presence of emerging contaminants in seafood;
• To better forecast climate change implications for seafood safety;
• To implement more mitigation strategies to minimize the risks of seafood contamination, including processing and phycoremediation;
• To improve communication with stakeholders, as a lack of trust and transparency still exists between consumers and authorities and the seafood industry;
• To improve consumers’ confidence in the seafood industry and authorities, transparent information is needed about product characteristics (e.g. origin, date of capture, safety guarantee) along the trade chain (i.e. producers, processors, distributors and retailers);
• Understanding consumer needs and perceptions is crucial to ensuring successful acceptance of seafood products;
• To use fast screening tools initially to detect contamination and then use conventional monitoring methodologies as a confirmatory measure. This will reduce costs and ensure faster and reliable results. It can be broadly used by food safety authorities and industry. These tools can be used by the industry as part of the internal quality assurance procedures (HACCP);
• To include new steps (e.g. heating) when assessing the levels of contaminants in seafood in order to get realistic information for consumers.

USEFUL TOOLS FOR SAFE SEAFOOD POLICY

FISHCHOICE: New online tool to evaluate benefits and risks of seafood consumption

The ECsafeSEAFOOD project partners have developed an online tool that balances the benefits and risks associated with seafood consumption, taking the exposure to contaminants into consideration. The tool is called FISHCHOICE and is available at: www.fishchoice.eu

Professional access can be requested online.
WHERE TO FIND ADDITIONAL INFORMATION

• Please visit the ECsafeSEAFOOD project website to view the project’s relevant deliverables (D2.4, D2.3, D2.7): www.ecsafeseafood.eu

• The EU REACH Regulation (Registration, Evaluation, Authorisation and Restriction of Chemical substances) was introduced in 2007 to improve the protection of human health and of the environment through better and earlier identification of the intrinsic properties of chemical substances. Under the REACH regime, manufacturers and importers are required to gather information on the properties of their chemical substances and to register the information. By registering and limiting the use of dangerous chemicals in the industrial production chain, REACH will indirectly reduce the contamination of marine waters and marine organisms by toxic substances. This will in turn decrease the risk of contamination of fish and seafood for human consumption. http://eur-lex.europa.eu/legal-content/EN/TXT/?uri=CELEX:32006R1907

• The EU’s Rapid Alert System for Food and Feed (RASFF) is a key tool to ensure the cross-border flow of information to swiftly react when risks to public health are detected in the food chain. RASFF enables information to be shared efficiently between its members (EU-28 national food safety authorities, European Commission, European Food Safety Authority, EFTA Surveillance Authority, Norway, Liechtenstein, Iceland and Switzerland) and provides a round-the-clock service to ensure that urgent notifications are sent, received and responded to collectively and efficiently. For more information or to access the RASFF portal, see: http://ec.europa.eu/food/safety/rasff_en

• FDA Fish and Fishery Products. Hazards and Controls Guidance (Fourth Edition) - April 2011. These guidelines are intended to assist processors of fish and fishery products in the development of their Hazard Analysis Critical Control Point (HACCP) plans. The guidelines will help processors of fish and fishery products to identify hazards that are associated with their products and formulate control strategies. http://www.fda.gov/downloads/Food/GuidanceRegulation/UCM251970.pdf


• EFSA Statement on the benefits of fish/seafood consumption compared to the risks of methylmercury in fish/seafood: https://www.efsa.europa.eu/en/efsajournal/pub/3982


WHERE TO FIND ADDITIONAL INFORMATION
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ECsafeSEAFOOD has been assessing food safety issues related to priority contaminants present in seafood as a result of environmental contamination (including those originating from harmful algal blooms and those associated with marine litter) and evaluating their impact on public health.

Availability of safe and high-quality food is a growing public concern and research plays a very important role in ensuring consumer confidence in this sector. ECsafeSEAFOOD will provide scientific evidence to serve as a basis for further development of common food safety, public health, and environmental policies and measures, by seeking to establish a quantitative link between the contamination of the marine environment and that of seafood.

ECsafeSEAFOOD aims to increase consumer confidence through the provision of clear and practical communication and information, working in close collaboration with food safety authorities.

For more information, please visit the project website, www.ecsafeseafood.eu, or contact the project coordinator, Dr. António Marques, amarques@ipma.pt.

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This guide is not intended as a comprehensive seafood safety guide. It is designed to provide information to assist decision-making regarding seafood safety. It has been developed based on scientific evidence from the project, as well as the best available evidence at the time of development of this publication.