Network of reference laboratories for monitoring of emerging environmental pollutants

NEWSLETTER

http://www.norman-network.net

WELCOME TO ISSUE N°3
OF THE NORMAN NETWORK NEWSLETTER

The aim of the activities of the NORMAN network is to enhance the exchange of information on emerging environmental substances, and to encourage the validation and harmonisation of common measurement methods and monitoring tools so that the requirements of risk assessors and risk managers can be better met. The NORMAN newsletter is for everyone interested in emerging substances in the environment. This newsletter keeps you up to date on scientific advances in this area and highlights the activities and events of the EU NORMAN Network.

Editorial

Protection of the Danube river: what can bring NORMAN network?

Igor LISKA
Secretariat of the International Commission for the Protection of the Danube River
e-mail: igor.liska@unvienna.org

The implementation of the EU Water Framework Directive is a top priority issue for the International Commission for the Protection of the Danube River (ICPDR). The WFD sets the environmental objectives, which include achieving a good chemical status and a good ecological status. The assessment of the chemical status and ecological status necessitates having at hand a battery of reliable and robust analytical methods providing accurate and precise data on the Priority Substances (identified as pollutants of concern at European level) and other pollutants (Specific Pollutants in the River Basins).

Current monitoring programmes under the ICPDR include the regular monitoring of a limited number of heavy metals and organic micropollutants within the Transnational Monitoring Network. On top of that, a full set of WFD priority substances and some emerging substances are monitored within the longitudinal surveys known as the Joint Danube Survey. Even though such a "single shot" survey cannot replace monitoring programmes required by the WFD, it represents a unique opportunity for a thorough screening for a wide variety of chemical substances, especially in areas with limited possibilities of state-of-the-art environmental trace analysis. The second Joint Danube Survey (JDS2) was organised in August – September this year and it is described in greater detail elsewhere in this Newsletter. JDS2 reconfirmed the need to intensify efforts to strengthen analytical capacities in the Danube River Basin as, in future, it is inevitable that the analysis of a wide range of priority and other substances will be an issue of common interest in all the Danube countries and that these compounds will be monitored regularly in each sub-basin. The process of upgrading the capacities of the water laboratories will primarily require substantial technical and financial support at the national level, but progress will also require an intensive information exchange and international cooperation. The planned NORMAN network could be in a position to contribute to that developing process by providing the necessary background information as well as guidance on analytical tools including QA/QC. This would be especially helpful in support of the national regulatory processes for the Specific Pollutants in the River Basins. Through the network the water laboratories could have access to information on key issues such as:

- Sampling procedures and analytical methodologies for emerging substances;
- Information and respective recommendations for internal QA/QC procedures compatible with the standardized accreditation requirements;
- Interlaboratory proficiency testing scheme (round robin tests);
- List of available/proposed national environmental quality standards for water, suspended particulate matter, sediments and biota;
- Experience with application of ecotoxicological analyses and their combination with the results of chemical screening methods;
- General guidance on analysis of emerging substances.

JDS2 not only contributed to greater insight into the occurrence of priority substances in the Danube River Basin, but also strengthened the basin-wide network of expertise in emerging substances. The planned NORMAN network, with its ambitious goals operating across a much larger geographical area, could further contribute to an upgrade of analytical skills necessary for WFD-compliant chemical monitoring.

NORMAN – Contract N° 018486 – is a Co-ordination Action funded by the European Commission, DG Research, under the FP6 – Priority 6.3 " Global Change and Ecosystems "

Issue 3 – December 2007

EDITORIAL p1
MONITORING AND BIO-MONITORING p2
ENVIRONMENTAL AND HUMAN HEALTH RISK ASSESSMENT p5
QUALITY ASSURANCE & QUALITY CONTROL p9
RESEARCH PROJECTS / FINDINGS p10
LIFE OF THE NETWORK p17
CALENDAR OF EVENTS p18

Monitoring and Bio-monitoring

Triclosan in plasma and milk from Swedish nursing mothers and their exposure via personal care products

In the past few years, the bactericidal compound triclosan has been the subject of various collaborative studies carried out by research groups from Stockholm University and the Karolinska Institutet. Triclosan is commonly used in e.g. plastics, textiles and health care products such as tooth pastes and deodorants.

The present study describes the findings of the determination of triclosan in plasma and breast milk of a relatively small (n=36) group of nursing mothers. The analytical methodology used to accurately quantify triclosan levels, including an acidic hydrolysis step to release any triclosan bound in conjugates, had been published earlier (Allmyr et al., 2006) and was applied while taking quality control parameters such as method repeatability into account.

It was shown that triclosan and/or its metabolites are omnipresent in both plasma and breast milk from nursing mothers and that concentrations in both matrices were clearly and significantly higher in the exposed group (i.e., the group that uses products containing triclosan) than in the control group. For both matrices there is a large within-group variability. Furthermore, the triclosan concentration in milk was lower than in plasma on an individual basis, which is influenced by the presence and behaviour of triclosan conjugates in plasma and milk and other factors such as protein binding and partitioning to lipids in either matrix. In any case, the infant is exposed to a considerably smaller dose of triclosan via the breast milk compared to the dose in the mother. The authors state that for the exposure of infants, direct contact with products that contain triclosan may be more important.

The significance of the presence of triclosan in plasma and breast milk of nursing mothers in terms of health effects is not easily deduced: long-term effects of chronic exposure to triclosan are not fully understood. In the paper, several triclosan-enyzme reactions are briefly discussed, such as the capacity to inhibit the iodothyronine hormone sulfotransferase activity in rat liver cytosol in vitro (Schuur et al., 1998). Summarizing, the findings suggest that the possibility cannot be excluded that triclosan exerts adverse effects on biological systems by interfering with the biotransformation of other exogenous and endogenous compounds.

It should be taken into account that triclosan levels in plasma and in breast milk may vary from country to country, possibly influenced by the advice on the use of products containing disinfectants in relation to common hygiene measures that have been formulated by national authorities such as in Sweden and The Netherlands (Health Council of the Netherlands, 2001). However, the general presence of triclosan in plasma from individuals who do not use products containing this compound indicates that there are other significant sources of exposure. For example, the fact that triclosan has been found in fish bile by applying Effect Directed Analysis (Houtman et al., 2004) clearly shows that triclosan has entered our environment in measurable quantities.

REFERENCES

- Allmyr M, McLachlan MS, Sandborgh-Englund G and Adolfsson-Erici M. ***Determination of triclosan as its pentaflurobenzoyl ester in human plasma and milk using electron capture negative ionization mass spectrometry***. Analytical Chemistry 78 [2006]: 6542-6546.
- Health Council of the Netherlands: ***Disinfectants in consumer products***. The Hague: Health Council of the Netherlands, 2001; publication no. 2001/05.
- Houtman CJ, van Oostveen AM, Brouwer A et al. ***Identification of estrogenic compounds in fish bile using bioassay-directed fractionation***. Environmental Science&Technology 38 [2004]: 6415-6423.
- Schuur AG, Legger FF, van Meeteren ME et al. ***In vitro inhibition of thyroid hormone sulfation by hydroxylated metabolites of halogenated aromatic hydrocarbons***. Chemical Research in Toxicology 11 [1998]: 1075-1081.

SOURCE:

Allmyr M, Adolfsson-Erici M, McLachlan MS, Sandborgh-Englund G. ***Triclosan in plasma and milk from Swedish nursing mothers and their exposure via personal care products***. Science of the Total Environment 372 [2006]: 87-93.

REVIEWED BY:

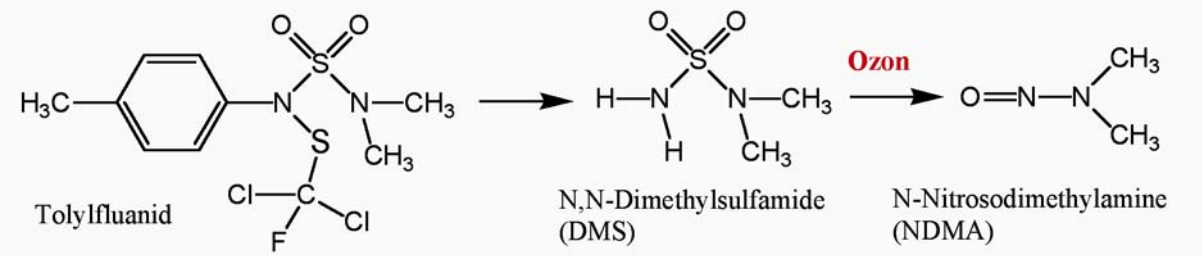
Marja LAMOREE
Institute for Environmental Studies, VU University, Amsterdam, NL
marja.lamoree@ivm.vu.nl

NORMAN – Contract N° 018486 – is a Co-ordination Action funded by the European Commission, DG Research, under the FP6 – Priority 6.3 " Global Change and Ecosystems "

A new fungicide metabolite

New research shows that N,N-Dimethylsulfamide (DMS), a hitherto unknown metabolite of the important fungicide Tolylfluanid, can form, by ozonation in water works, the human carcinogen N-Nitroso-dimethylamine (NDMA).

Dr. Carsten Schmidt from the German "Technologie Zentrum Wasser" TZW in Karlsruhe observed in autumn 2006 the formation of N-Nitro-sodimethylamine (NDMA), a human carcinogen, in water-works during ozonation treatment. It was found that NDMA is formed from DMS, an unknown metabolite of Tolylfluanid.



Tolyfluanid is a fungicide applied for the control of fungus diseases in apple, grape, hop, and tomato production. The German Federal Office for consumer protection and food safety, acting on the precautionary principle, withdrew the authorisation for pesticide products containing Tolylfluanid on 21 February 2007 [1] and a drinking water plant with NDMA levels of ~ 200 ng/L was closed. In the USA a drinking water action level of 20 ng/L has been set in the past [2].

During drinking water treatment, DMS is difficult to remove by floccu-lation, UV disinfection or activated carbon filtration, and is therefore classified as a drinking water-relevant compound. By ozonation, approximately half of the DMS-molecules form NDMA (CAS 62-75-9). In oxidation with H_2O_2, chlorine or chlorine dioxide, DMS is degraded to unknown degradation products, but not NDMA [1].

In the USA, Canada and Australia, NDMA has been known for some time to be a disinfection by-product of the chloramination process, which is applied in those countries, but not allowed in Germany. NDMA can form at approximately 10-100 ng/L during chlorination [2] of secondary wastewaters and chloramination of surface waters [3].

Several treatment methods for the degradation of NDMA in water, such as ultraviolet (UV) photolysis, or photocatalytic degradation with TiO_2, have been suggested. UV treatment is known to be the most efficient method due to the strong photolability of NDMA. It has been demons-trated that NDMA contaminated water can be effectively treated using low-pressure or medium-pressure Hg lamps [3].

Another method for controlling the NDMA level in water is the preven-tion of NDMA formation by removing NDMA precursors, which is a more fundamental strategy than degrading the NDMA once it is formed [4].

Analysis of NDMA is performed by solid-phase extraction (SPE) using a combination of C18 and spherical carbonaceous sorbent followed by an HPLC tandem mass spectrometry technique (MS-MS), employing high mass resolution, with positive electrospray ionization (ESI+) in the multiple reaction monitoring mode (MRM).

Drinking water should be free of carcinogenic compounds. This exam-ple shows that the authorisation of industrial chemicals should also include the investigation of their possibly toxic degradation products which might be formed during water purification processes in water-works.

REFERENCES

[1] Schmidt C. TZW Aktuell, ***Nachrichten aus dem Technologiezentrum Wasser***, Karlsruhe, DVGW, 22 May 2007, **http://www.tzw.de/pdf/TZWaktuell22.pdf**

[2] ATSDR, US Agency for Toxic Substances and Disease Registry **http://www.atsdr.cdc.gov/tfacts141.html**

[3] Mitch WA, Sedlak DL. ***Formation of N-Nitrosodimethylamine (NDMA) from Dimethylamine during Chlorination***, Environ. Sci. Technol. 36 [2002]: 588-595.

[4] Lee C, Schmidt C, Yoon J and von Gunten U. ***Oxidation of N-Nitrosodimethylamine (NDMA) Precursors with Ozone and Chlorine Dioxide: Kinetics and Effect on NDMA Formation Potential***, Environ. Sci. Technol. 41 [2007]: 2056-2063.

REVIEWED BY:

Robert LOOS
Institute for Environment and Sustainability, Joint Research Centre (JRC), Ispra, Italy
robert.loos@jrc.it

NORMAN – Contract N° 018486 – is a Co-ordination Action funded by the European Commission, DG Research, under the FP6 – Priority 6.3 " Global Change and Ecosystems "

Occurrence of polycyclic musks in sewage sludge and their behaviour in soils and plants

Synthetic musk fragrances are a group of chemicals used in a wide range of applications (detergents, perfumes, shampoos, and other personal care products). Synthetic musks have been used for many decades and the specific use of polycyclic musks has particularly increased in the last decade. The most widely used polycyclic musks are HHCB (Galaxolide®) and AHTN (Tonalide®). These two products account for 95% of the total market volume for polycyclic musks (PCMs) and are the most frequently detected PCMs. The other PCMs are ADBI (Celestolide®), ATTN (Versalide®), AHDI (Phantolide®) and ATII (Traseolide®).

Because of the relatively high lipophilicity of these compounds, they can accumulate in different matrices. PCMs have been detected in sewage sludges, surface waters, sediments, aquatic organisms and other biota. Transport in the water pathway is mainly supported by suspended particulate matter and can occur over long distances.

The first paper (Part 1) presented analytical developments for the determination of HHCB, AHTN, ADBI, ATTN, AHD and ATII in sludge samples and application to 21 sewage treatment plants with sampling in summer and winter. The authors showed the presence of PCMs, with concentrations ranging from 2.9 to 10.4 mg/kg in activated sludge samples and concluded that PCMs and particularly HHCB and AHTN should be considered in a risk assessment as potential contaminants of sewage sludge destined for agricultural use. According to the authors, important seasonal variations occurred and are linked to changes in degradation and volatilisation phenomena. These first results indicate that PCMs can be transferred to agricultural soils by the use of sewage sludge as a fertiliser.

The aim of the second paper (Part 2) was to perform studies concerning the adsorption of PCMs to soil, their dissipation and leaching in soil and their uptake by plants. As major detected compounds in sewage sludge, HHCB and AHTN were selected for experiments. Soil samples having different properties in organic carbon content, in pH and clay content were selected for sorption, leaching, dissipation and plant uptake studies.

Sorption kinetic studies showed that PCMs were adsorbed in a very short time and more strongly by the high organic carbon content soil. The authors calculated and used the Freundlich adsorption coefficient (Kf) and normalised sorption constants (Koc) to compare the adsorption behaviour of compounds in different soils. The organic matter content influenced adsorption more efficiently than the clay content. The adsorption rates ranged from 75.4 to 95.4% depending on the soil, and are comparable to those measured for flame retardants or PAHs. During the dissipation studies, slow aerobic dissipation and a low degradation rate were measured at 50% for HHCB and 27.5% for AHTN respectively, allowing the authors to extrapolate these half-life times for dissipation: 10-17 months for HHCB and 2-14 years for AHTN. No relevant microbial degradation, low abiotic degradation or elimination by volatilization took place.

Leaching studies demonstrated the low mobility of PCMs, with no enhancement linked to dissolved organic carbon content. The experiment showed leaching rates <0.001% for HHCB and AHTN during a test period of 48 h.

Plant uptake studies were carried out on lettuce and carrots (leaves and roots) and showed low contamination of lettuce plants (max. concentration: 290 µg/kg for HHCB and 820 µg/kg for AHTN after 6 weeks, followed by a decrease to 100 µg/kg and 440 µg/kg respectively after 12 weeks). Uptake studies with carrots resulted in very high concentrations in carrot roots. Depending on the tested soil, values from 2mg/kg to 14mg/kg were measured after 18 weeks, following a contamination peak observed after 14 weeks (ranged from 4 to 20 mg/kg). The authors suggested that the occurrence of surfactant in sewage sludge (not measured in this study, but frequently detected in this kind of sample) can facilitate uptake by carrots and that the occurrence of essential oil cells in carrot roots can increase the accumulation capacity of this vegetable. In the worst case, the calculated transfer factor reached 0.48.

To conclude, the authors consider that PCMs should be considered in risk assessment studies as a potential contaminant of sewage sludge. The application of the sludge to agricultural soil and the high persistence of these compounds can be involved in the contamination of the upper part of the soil and the potential contamination of vegetables such as carrots. These results highlight the need for further investigations to gain a better understanding of PCM plant uptake and to develop qualitative risk assessment studies in relation to the consumption of contaminated products.

SOURCES:

Muller J, Bohmer W and Litz NT. ***Occurrence of polycyclic musks in sewage sludge and their behaviour in soils and plants***. Journal of Soils and Sediments 6(4) [2006]: 231-235.

Litz NT, Muller J and Bohmer W. ***Occurrence of polycyclic musks in sewage sludge and their behaviour in soils and plants - Part 2: Investigation of polycyclic musks in soil and plants***. Journal of Soils and Sediments 7(1) [2007]: 36-44.

REVIEWED BY:

Anne TOGOLA
BRGM Metrology, Monitoring and Analysis, France
a.togola@brgm.fr

NORMAN – Contract N° 018486 – is a Co-ordination Action funded by the European Commission, DG Research, under the FP6 – Priority 6.3 " Global Change and Ecosystems "

PFOS and PFOA in plasma of German population

In recent years, perfluorinated compounds (PFCs) have been found in the environment (water, biota, air, sludge) and in human media such as plasma and blood. Fromme et al. and Midasch et al. report on perfluorooctane sulfonate (PFOS) and perfluorooctanoic acid (PFOA) levels in human plasma samples. These samples originate from Bavarian adults and the German general population.

The method of analysis used here has an interesting feature. In most published methods, analysis starts with denaturation of plasma, serum or blood proteins and removal by centrifugation. This is then followed by a solid phase extraction (SPE) for preconcentration and clean-up of samples and analysis by LC-ESI-MS/MS. These authors took out the SPE part and basically directly injected the sample in the LC-ESI-MS/MS (after denaturation of proteins). In doing so, they obtained satisfactory quantification limits and the accuracy and precision showed that their methods were fit for purpose.

The results of their studies show that the levels in plasma are 22 ng/mL for PFOS and 6.8 ng/mL for PFOA for the general German population, whereas the exposure of Bavarian population is lower (1 and 4.8 ng/mL, respectively). These levels correspond to those observed in other studies on non-occupationally exposed people. Midasch et al. found that (unlike lipophilic POPs), there is no build-up of concentrations with age, whereas Fromme et al. found a small, but significant increase with age (females only). The latter group also found increased concentrations to be associated with males (10-30% higher levels than females) and rural residence (2.5% increase compared to urban residence, PFOA only). Finally, a strong correlation between PFOA and PFOS concentrations ($r = 0.82$) suggest that the sources for both substances are similar.

These and other data show that PFCs behave differently from the lipophilic POPs. This calls for more efforts to asses their fate. Only limited information is available on PFCs in drinking water, food from vegetable and animal origin, food packaging, indoor and outdoor air and dust. The relevance of these sources in relation to human exposure to PFOS and PFOA remains to be elucidated.

SOURCES:

Fromme H, Midasch O, Twardella D et al. ***Occurrence of perfluorinated substances in an adult German population in southern Bavaria***. International Archives of Occupational and Environmental Health 80(4) [2007]: 313-319.

Midasch O, Schettgen T and Angerer J. ***Pilot study on the perfluorooctanesulfonate and perfluorooctanoate exposure of the German general population***. International Journal of Hygiene and Environmental Health 209(6) [2006]: 489-496.

REVIEWED BY:

Stefan VAN LEEUWEN
IVM - Institute for Environmental Studies, Vrije Universiteit, Amsterdam
stefan.van.leeuwen@ivm.falw.vu.nl

Environmental and human health risk assessment

Quantitative structure-retention relationships of pesticides in reversed-phase high-performance liquid chromatography

Estimation methods for physico-chemical properties of emerging compounds can be valuable tools for various purposes, including risk assessment, priority setting, quick response in case of emergencies, etc. Thereupon, in case of biological properties, numerical estimation methods may serve in reducing animal testing. Various estimation methods for a wide variety of endpoints have been reported so far. In this paper, Dr Aschi and co-workers report on the development of a novel method for estimating retention properties of pesticides in reversed-phase high-performance liquid chromatography (HPLC). HPLC is the technique of choice for relatively polar compounds such as the new generations of pesticides. As alternatives to traditional trial-and-error approaches, estimation methods for retention of organic pesticides are valuable tools in efficient monitoring campaigns.

NORMAN – Contract N° 018486 – is a Co-ordination Action funded by the European Commission, DG Research, under the FP6 – Priority 6.3 " Global Change and Ecosystems "

wo models are reported here: one based upon a six-parameter multiple linear regression model and a model derived by means of an artificial neural network analysis with back-propagation learning rules. As a matter of course, the artificial neural network-based model performs better than the multiple linear regression model (with estimates of retention properties as accurate as within 10 % in the case of the neural network-based model, versus a typical standard error of over 20 % in the case of multiple linear regression). Both models are thus well-suited to provide reliable estimates of retention times of pesticides, depending on the molecular structure and the solvent used. A major disadvantage of the neural network-based model, is, however, the mere fact that the model is available only to the developers of the model!

SOURCE:

Aschi M, D'Archivio AA, Maggi MA, Mazzeo P and Ruggieri F. Quantitative structure-retention relationships of pesticides in reversed-phase high-performance liquid chromatography. Analytica Chimica Acta 582(2) [2007]: 235-242.

REVIEWED BY:

Willie PEIJNENBURG
RIVM - The Netherlands National Institute for Public Health and the Environment
WJGM.Peijnenburg@rivm.nl

Unidentified inert ingredients in pesticides:
Implications for human and environmental health

Pesticides are a special class of toxic chemicals: unlike other chemicals, they are designed to kill, repel, or otherwise harm living organisms. They are also one of the few toxic substances that are intentionally applied to the environment. Pesticide ingredients are, by statute or by regulation, divided into two categories: active and so-called "inert" ingredients. The latter suggests that these ingredients are physically, chemically or biologically inactive. In fact, many consumers have the misleading impression that inert ingredients mean water or other harmless ingredients. Inert ingredients may constitute a significant proportion of typical pesticide formulations, with common household products containing over 86 % inert ingredients and >50 % for agricultural products.

Inert ingredients are added to act as solvents, surfactants or preservatives (among many other functions). A single product may contain a number of inert ingredients, each with a different purpose in the formulation. There is no requirement to identify inert ingredients on the label; it is claimed by pesticide manufacturers that some inert ingredient information is protected as confidential business information. Despite their name, inert ingredients may be biologically or chemically active and are labelled inert only because of their function in the formulated product:

1. Inert ingredients can increase the toxicity to humans of pesticide formulations. Studies with and without inclusion of inert ingredients clearly showed more severe effects when inert ingredients were included, albeit that most studies were carried out in vitro;
2. Inert ingredients can increase exposure to pesticide formulations. Inert and active ingredients can, for example, interact to diminish the protective efficacy of clothing and skin, reduce the efficacy of washing, and increase persistence and off-target movement of pesticides;
3. Inert ingredients can increase ecotoxicity of pesticide formulations: the severity of toxic effects of active ingredients in pesticides in non-target plants, animals, and microorganisms can be increased by the inert ingredients with which they are formulated. The most common example is glyphosphate, with some formulations being 10-100 times more acutely toxic to fish than is the active ingredient alone.

On the basis of the description of increased effects of inert ingredients, Cox and Surgan conclude that pesticide registration should consistently require full impact assessment of formulations, whereas environmental monitoring should increasingly include inert ingredients. Finally, it is advocated that despite confidentiality issues, inert ingredients should be identified on product labels. Chemical mixtures have recently been identified by the US Agency for Toxic Substances and Disease Registry as one of six priority areas in public health research. Health and environmental researchers should therefore support this emerging issue by independent investigations, unhindered by the secrecy that shrouds the inert ingredients in pesticide products.

SOURCE:

Cox C and Surgan M. ***Unidentified inert ingredients in pesticides: Implications for human and environmental health***. Environmental Health Perspectives 114(12) [2006]: 1803-1806.

REVIEWED BY:

Willie PEIJNENBURG
RIVM - The Netherlands National Institute for Public Health and the Environment
WJGM.Peijnenburg@rivm.nl

NORMAN – Contract N° 018486 – is a Co-ordination Action funded by the European Commission, DG Research, under the FP6 – Priority 6.3 " Global Change and Ecosystems "

How to assess the risks of nanotechnology:
Learning from past experience

The design, production and use of materials and devices less than 100 nm in size are likely to expand (exponentially?) in the near future. Benefits from any new technological advance are usually so compelling that it may be difficult to contemplate its possible negative implications. This is even despite warnings issued at an early stage by experts from various disciplines. Proper risk assessment requires qualitative and quantitative information on the various facets that jointly determine the extent of adverse effects. An important factor here is that, while technocrats make initial decisions the public eventually decides which risks and benefits are acceptable. The power of public opinion in the face of technological innovation should not be ignored in this context. Examples such as the opposition to nuclear power generation and the public reaction to genetically modified crops and food are illustrative, highlighting the need to properly inform the public. Public perception may even be a factor in the attribution of health symptoms.

The rapidly expanding area of nanotechnology is no exception to the observation made above and efforts to develop and apply nanomaterials far exceed the research into the risks possibly associated with nanotechnology. There are at present many gaps in our knowledge of those properties of nanomaterials that may induce unexpected risks. Combined properties such as particle size, effective surface, hydrophobicity, potential of activating specific receptors of toxicity - and other risks that probably lie beyond our epistemological horizon - may induce currently unexpected risks to human health and to ecosystems. Some stakeholders have advocated strict application of the precautionary principle (place the burden of proof on those who have a commercial or other stake in developing the technological application) and even a provisional ban on nanomaterials. The precautionary principle provides little or no direction on how to increase knowledge of potential and actual risks; science might and often does! Accounts of lessons learned from previous environmental issues such as asbestos, PCBs, and methyl tertiary butyl ether (MTBE, an oxygenate added to fossil fuel to reduce emissions of precursor chemicals that contribute to the formation of tropospheric ozone) serve to illustrate the value of the precautionary principle and document problems that could have been predicted. The MTBE-case is especially illustrative, because key problems with MTBE not only could have been predicted but actually were anticipated. Quoting the adage that "history teaches us the mistakes we are about to make", Dr Davis uses the example of MTBE to show that we may be able to avert the most pessimistic predictions of the negative aspects of the rapidly emerging area of nanotechnology at an early stage.

The most important lessons to be learned from the MTBE-example include:

1. A systems approach towards risk assessment is needed, starting with a product life cycle perspective and including a multimedia environmental perspective;
2. By-products may be more problematic than the primary substance;
3. Human health is not the only issue of concern, and either the mere presence in groundwater or odour or taste may also catalyse concern. Other environmental issues, such as global warming and acid rain, may also overrule human health concerns;
4. Caution needs to be used when extrapolating from one species to another. In this case, tests performed with MTBE in rats were not predictive of effects on humans. The warning is especially applicable to nanomaterials as they may have unique toxicological properties that may vary with seemingly minor structural alterations;
5. Keep the public well-informed;
6. Everything has trade-offs, some of which are acceptable, while others are not. In the case of MTBE, the balance ultimately tipped to the negative side;
7. Technical experts may be able to anticipate risks, even with limited information;
8. An adaptive risk management strategy is critically important.

In view of these lessons, the author is advocating the proactive adoption of the strategy of "comprehensive environmental assessment" for nanomaterials, in which product life cycle analysis is combined with the risk assessment paradigm, taking the specific features of nanomaterials into account. Overall, the author is recognising that the problem with lessons learned is that they may seem trivially obvious in hindsight. Nevertheless, the conclusion of applying an adaptive risk management strategy in an iterative mode is quite useful to evaluate and even anticipate and mitigate environmental and human health risks associated with nanotechnology. Monitoring and research are to be combined with other sources of information to characterise risk and determine research priorities.

SOURCE:

Davis JM. ***How to assess the risks of nanotechnology: Learning from past experience***. Journal of Nanoscience and Nanotechnology 7(2) [2007]: 402-409.

REVIEWED BY:

Willie PEIJNENBURG
RIVM - The Netherlands National Institute for Public Health and the Environment
WJGM.Peijnenburg@rivm.nl

Expression of heat shock protein and hemoglobin genes in Chironomus tentans (Diptera, chironomidae) larvae exposed to various environmental pollutants: A potential biomarker of freshwater monitoring

A serious quest is going on for biomarkers of exposure to chemicals. Various approaches, with varying degree of success, have been advocated for various biota. Ideally, biomarker responses are directly linked to exposure to specific contaminants. However, in common practice it is difficult to establish a causality relationship between biomarker response and actual exposure, let alone that biomarkers may currently be used for water, soil or air quality monitoring.

Larvae of Chironomus tentans were used by Dr. Lee and co-workers to develop a sensitive biomarker for freshwater monitoring, based on pollutant-induced expression of heat shock proteins (HSPs) and hemoglobins (Hbs) genes by a diversity of chemical ranging from metals to surfactants and steroids. In addition, larvae growth was monitored as a physiological effect measure.

The authors conclude that response of the HSP gene expression was rapid and sensitive to low chemical concentrations, as opposed to physiological effects. Expression of HSP genes was, however, not chemical specific. Interestingly, the latter was indeed the case for Hb genes expression: whereas alkyl phenolic compounds increased expression of the hemoglobin genes, the expression was decreased by pesticides. Expression of Hb genes could be due to increase in oxygen demand for xenobiotic metabolisation processes. However, the observed lack of expression in case of pesticides cannot be accounted for.

These findings suggest that expression HSP and Hb genes in Chironomus could give useful information for diagnosing the general health condition of the freshwater ecosystem. However, monitoring strategies based upon induction of these genes are still one bridge too far given the non-specificity of the assays, the more since a limited number of chemicals were tested.

SOURCE:

Lee SM, Lee SB, Park CH and Choi J. ***Expression of heat shock protein and hemoglobin genes in Chironomus tentans (Diptera, chironomidae) larvae exposed to various environmental pollutants: A potential biomarker of freshwater monitoring***. Chemosphere 65(6) [2006]: 1074-1081.

REVIEWED BY:

Willie PEIJNENBURG
RIVM - The Netherlands National Institute for Public Health and the Environment
WJGM.Peijnenburg@rivm.nl

Ecotoxicological Risk of Pharmaceuticals from Wastewater Treatment Plants

The potential risks of pharmaceuticals as toxic contaminants in the aquatic environment have become a research focus only in the last few years. This article investigated the ecotoxicological effects of pharmaceutically active compounds (PhAC) detected in the effluents of Korean wastewater treatment plants on Daphnia magna, using biological and chemical analyses.

Daphnia magna were exposed to PhAC during 48-h acute toxicity tests and 21-d chronic reproduction tests either as single compounds or as mixtures. The selection of target pharmaceuticals was based upon their occurrence in wastewater effluents, their broad application and their potential ecological effects. Results were expressed as LC50 for the acute tests and NOEC for the chronic tests. Additionally, chemical analysis by GC-MS was performed on influents and effluents from major waste water treatment plants in South Korea.

The single compound toxicity tests indicated that most of the pharmaceuticals tested were toxic and harmful to aquatic organisms. However, the concentrations required to produce these effects (LC50 7.4-141.2 mg/L, NOEC 10-40 mg/L) were well above those chemically determined in the wastewater treatment effluents (0.06-10.96 µg/L). Furthermore, the study found that combined toxicity of PhAC mixtures displayed a small synergistic effect in both acute and chronic tests but not at concentrations that would be considered critically harmful.

Ecological risk assessments were performed to characterize the degree of contamination and to evaluate the adverse effects of these chemicals in the aquatic environment. Data from the toxicity tests was used alongside the GC-MS analyses of selected compounds in the influents and effluents in order to evaluate the degree of risk from each PhAC (Hazard quotient, HQ). The study employed an assessment factor of 1,000 as recommended by the EU and OECD. When the assessment value was applied, any HQ value less than 1 would indicate there was no significant risk. None of the HQ values of the pharmaceuticals tested exceeded 1, and the study concluded that no acute or chronic effects to D. magna were caused by the pharmaceuticals in Korean wastewater treatment effluents. The authors acknowledge that neither the volume of effluent discharged nor the dilution factor from the receiving water was considered in the risk assessment. This information is essential in assessing the actual environmental risk from an effluent and the actual impacts of the Korean wastewater treatment effluents upon Daphnia magna can therefore not be determined.

In conclusion, though the study showed no significant ecotoxicological effects, it does add to our limited scientific knowledge of the potential impacts of pharmaceuticals on the aquatic environment. Additionally, it identifies the need for further bioassay studies using algae, daphnids and fish exposed to pharmaceuticals and their derivatives identified in wastewater treatment effluents. Investigations must include invertebrate studies at the sub-lethal level (physiological, molecular and population measures) using environmentally realistic concentrations of pharmaceuticals in order for us to understand the mechanisms by which these chemicals elicit toxicity.

SOURCE:

Han GH, Hur HG and Kim SD. ***Ecotoxicological risk of pharmaceuticals from wastewater treatment plants in Korea: Occurrence and toxicity to Daphnia magna***. Environmental Toxicology and Chemistry 25(1) [2006]: 265-271.

REVIEWED BY:

Anne O'NEILL
UKEA – The Environment Agency, United Kingdom
anne.oneill@environment-agency.gov.uk

NORMAN – Contract N° 018486 – is a Co-ordination Action funded by the European Commission, DG Research, under the FP6 – Priority 6.3 " Global Change and Ecosystems "

Determination of pharmaceuticals from different therapeutic classes in wastewaters

Pharmaceuticals in the environment is a subject of growing scientific and societal concern, and there is a need to carry out evaluations of human and ecological risks associated with the presence of these complex mixtures of organic micropollutants in water resources. Exposure assessment is the backbone of these risk assessments, and requires reliable analytical methods.

In this context, Botisi et al. describe an analytical method for simultaneous determination of pharmaceuticals from different therapeutic classes in effluent wastewaters. Target molecules have partially been chosen with the aid of the European Surveillance of Antimicrobial Consumption data (http://www.ua.ac.be/ESAC) that could be seen as an interesting prioritisation tool – reporting reliable human antibiotic use statistics – for European authors interested in the topic of antibiotics in the environment. In this article seven pharmaceuticals, widely used in human and veterinary medicine, are selected: five sulfonamides and trimethoprim antimicrobials and the non-steroid anti-inflammatory agent diclofenac.

The analytical method itself could be seen as a classical successful development of an off-line solid phase extraction (SPE) followed by reverse-phase liquid chromatography with triple quadrupole mass spectrometry (LC/MS/MS). Additional developments, significantly increasing confidence in the reported results and the method, are, however, presented in two complementary areas: identification of the target molecules and control of the signal suppression. For the identification of the target molecules, an application of the EU Commission Decision 2002/657/EC – developed for the monitoring of residues in products of animal origin – as already reported for pharmaceuticals in the environment, is presented. With the application of the LC/MS/MS protocol with 1 precursor ion (low mass resolution) and 2 daughters with a specific ratio, 4 identification points (IP) are earned, compared with the minimum three identification points required by EU for a confirmatory method. As a consequence of this QA/QC need, method detection and quantification limits (MDL and MQL) in the final matrix could be slightly increased by the fact that they are calculated for the less intense transition. Nevertheless, in this article, MQL for effluent wastewaters were in the range 12-20 ng.L-1 probably achieved because of the selection of a limited peak-width value (0.2 Da) on the first mass analyzer (Q1). For the control of the signal suppression, a comparison based on the signal intensity from each target molecule in the sample matrix compared with that in deionised water was performed. Despite the application of a variety of classical measures to limit the signal suppression (sample dilution in deionised water, flow rate of 0.2 mL.min-1, use of a single isotopically labelled internal standard), the reduction of responses is reported to be in the range 15-35%. As a consequence, quantification of target molecules in real samples was performed using the standard addition method, except for the compound corresponding to the internal standard. Method accuracy and precision were evaluated by recovery studies at three concentrations determined in triplicate on three different days.

Concentrations in final effluents of Greek wastewater treatment plants (WWTP) after biological treatment are reported. Sulfamethoxazole, trimetoprim and diclofenac have been quantified at concentrations from 50 to 400 ng.L-1 with higher concentrations in the WWTP receiving hospital wastewater. The last figure of note in this article is that monitored effluents were collected as composite samples, thus data provided in concentration could easily be converted in mass flows. It is the reviewer's point of view that the quality and utility of low level concentrations reported in complex matrices with the use of SPE-LC/MS/MS methods mainly depend on explanations provided about the four following critical points: selection of target molecules, sampling strategy, identification of the target molecules and management of matrix effects (among them signal suppression, especially for LC/MS methods).

FURTHER READING:

- ***EU Commission Decision 2002/657/EC*** of 12 August 2002 implementing Council Directive 96/23/EC concerning the performance of analytical methods and the interpretation of results (available at: http://eur-lex.europa.eu/LexUriServ/site/en/oj/2002/l_221/l_22120020817en00080036.pdf)
- Petrovic M, Gros M & Barcelo D. ***Multi-redisue analysis of pharmaceuticals in wastewater by ultra-performance liquid chromatography-quadrupole-time-of-flight mass spectrometry***. Journal of Chromatography A 1124 [2006]: 68-81.
- Gomez MJ, Martinez Bueno MJ, Lacorte S et al. ***Pilot survey monitoring pharmaceuticals and related compounds in a sewage treatment plant located on the Mediterranean coast***. Chemosphere 66 [2006]: 993-1002.

SOURCE:

Botitsi E, Frosyni C and Tsipi D. ***Determination of pharmaceuticals from different therapeutic classes in wastewaters by liquid chromatography-electrospray ionization-tandem mass spectrometry***. Analytical and Bioanalytical Chemistry 387(4) [2007]: 1317-1327.

REVIEWED BY:

Jean-Ulrich MULLOT
Laboratoire Santé Publique – Environnement. Université Paris Sud 11, Faculté de Pharmacie de Chatenay-Malabry, France
jumullot@aol.com

NORMAN – Contract N° 018486 – is a Co-ordination Action funded by the European Commission, DG Research, under the FP6 – Priority 6.3 " Global Change and Ecosystems "

Total fluorine in water and in human blood

In these two papers, Miyake and co-workers employ a novel analytical approach to estimate the total quantity of extractable organic fluorine (EOF) in seawater and human blood samples. The same samples were also analysed for known perfluorinated compounds such as PFOS and PFOA. By comparing the total fluorine content and the EOF with the "known" organic fluorine, the authors estimate the quantity of unknown organic fluorine in these matrices. This provides interesting insight into the existence of further as yet unknown chemicals in this important group of emerging contaminants.

The major analytical innovation in these papers is the development of a method for extractable organic fluorine (EOF) that is sensitive and selective enough to allow quantification in these matrices. The instrumental method is based on combustion of the sample at 900-1000 °C to convert organic fluorine into hydrogen fluoride, which is then trapped in a sodium hydroxide solution and analysed using ion chromatography. Prior to instrumental analysis, the seawater samples were first extracted on an Oasis®WAX SPE cartridge and then eluted with methanol and 0.1 % NH_4OH / methanol. Since the fluoride ions were also retained on this sorbent, a method was developed to chromatographically separate the fluoride from the organic fluorinated chemicals by washing the SPE cartridge with 0.01 % NH_4OH / H_2O. Good recoveries (81-109 %) are reported for known perfluorinated chemicals (PFCs), and the authors claim that the method allowed quantification of EOF at the sub-µg/L level, but documentation of the LOQ determination and method reproducibility is missing. For the blood samples a different approach was taken. An ion pair extraction with MTBE and hexane was employed. The recoveries of the known PFCs using this extraction method were again good, and in this case the reproducibility (CV of 4-10 %), and LOD (3 µg-F/L) are reported.

Two "background" seawater samples were analysed using these methods. Both yielded EOF values of ~100 ng-F/L. The analysed PFCs (the C4-, C6- and C8-perfluorosulfonates, the C5- through C11-perfluorocarboxylates, and PFOSA) accounted for just 1% and 2.5% of the EOF. This indicates that there are many other organic fluorine compounds present in seawater. The authors suggest that fluorinated acetic acids, and in particular trifluoroacetic acid (TFA), may make up a major portion of the EOF, and highlight the need for measurements of these compounds in seawater to better constrain the levels of unknown fluorinated organic chemicals in seawater. However, it is not clear whether these compounds were efficiently extracted on the Oasis®WAX SPE cartridge, so the origin of the unknown fluorinated chemicals remains speculative.

In "background" whole blood samples from Japan and plasma samples from the USA, EOF values of <6-9 and 18-59 ng-F/mL, respectively, were measured. In this case the known PFCs accounted for almost all of the EOF, with PFOS contributing the majority in both countries. It should be noted that the ion pair extraction method used for the EOF determination is extremely specific for the known PFCs, and this may explain why there was good agreement between the EOF and PFC determinations. However, the total fluorine was 3-20 times greater than the EOF in these samples. The authors could not distinguish between inorganic fluoride and non-extractable organic fluorine, and they speculate that the samples may have contained several unidentified non-extractable organic forms of fluorine.

In summary, the authors do not provide us with a quantitative answer regarding the presence of unknown fluorinated organic chemicals in the environment. However, they do offer analytical methods and a mass balance methodology that will be useful tools in further exploring this issue.

SOURCES:

Miyake Y, Yamashita N, Rostkowski P et al. ***Determination of trace levels of total fluorine in water using combustion ion chromatography for fluorine: A mass balance approach to determine individual perfluorinated chemicals in water***. Journal of Chromatography A 1143(1-2) [2007]: 98-104.

Miyake Y, Yamashita N, So MK et al. ***Trace analysis of total fluorine in human blood using combustion ion chromatography for fluorine: A mass balance approach for the determination of known and unknown organofluorine compounds***. Journal of Chromatography A 1154 [2007]: 214-221.

REVIEWED BY:

Michael MCLACHLAN
ITM - Department of Applied Environmental Science, Stockholm University, Stockholm, Sweden
michael.mclachlan@itm.su.se

Research projects / findings

Joint Danube Surveys

ICPDR tool for searching the emerging substances

Co-ordinator Igor LISKA
International Commission for the Protection of the Danube River, Vienna International Centre,
D 0412, P.O.Box 500, 1400 Vienna, Austria
http://www.icpdr.org/jds/

Water pollution is one of the key problems of the Danube River Basin. In response, a basin-wide monitoring network, the so-called Transnational Monitoring Network (TNMN), was established under the International Commission for the Protection of the Danube River (ICPDR) and has been in operation since 1996. However, given the size and complexity of the Danube River Basin, this network focuses only on a standard set of chemical and biological parameters.

To get a better insight into water quality, especially the occurrence of specific dangerous substances or a thorough hydrobiological characterisation, complementary monitoring activity has been established in the Danube River Basin, and is achieved via scientific longitudinal surveys on the whole stretch of the river. The first Joint Danube Survey (JDS1) was carried out in 2001 and its results were a key information source for characterisation of the Danube River Basin District as required by the EU WFD.

NORMAN – Contract N° 018486 – is a Co-ordination Action funded by the European Commission, DG Research, under the FP6 – Priority 6.3 " Global Change and Ecosystems "

Among large sets of chemical and biological parameters, JDS 1 focused on the monitoring of para-tert-octylphenol, 4-iso-nonylphenol, di[2-ethylhexyl]phthalate, pentachlorophenol, pentabromodiphenyl ether and tributyltin in suspended particulate matter and sediment samples as well as of selected pharmaceuticals in water from the river Danube and its major tributaries. No pentabromodiphenyl ether or pentachlorophenol was detected in any of the JDS1 samples, and tributyltin was found in relatively low concentrations. Para-tert-octylphenol was detected exclusively in bottom sediments, while 4-iso-nonylphenol and di[2-ethylhexyl]phthalate were found in bottom sediments as well as in suspended solids in significant concentrations (from a few µg/kg up to more than 100 mg/kg), indicating the relevance of these compounds as markers for industrial pollution of solids in the river Danube. The results for pharmaceuticals showed that only a few of the analytes extracted at neutral pH were detectable. The only substances present in nearly every sample were isopropylphenazone, which was found in low concentrations of 0.003-0.004 µg/l, and N-acetyl-4-aminoantipyrine (metabolite of metamizol), whose concentrations showed larger variations. From Budapest downstream, the concentrations of N-acetyl-4-aminoantipyrine showed a relatively constant level of around 0.1 µg/l. Three samples showed significantly higher amounts of this substance, namely Iskar (Sofia discharge), Jantra and Arges (Bucharest discharge). Another metabolite of metamizol, N-formyl-4-aminoantipyrine, showed the same behaviour, but could not be detected in some cases due to a too high limit of detection. The three samples mentioned above also contained the analgesic phenazone in concentration range 0.029 - 0.46 µg/l.
In addition to the target analyses of organic substances, a GC-MS screening of water and sediment samples was performed during JDS1. Altogether 98 water samples were analysed by GC-MS and in each sample a number of organic compounds were identified. Based on the spectral information collected, the chemical structures of 96 compounds occurring in the Danube could be suggested. The most ubiquitous compounds in water were phthalates, fatty acids, aliphatic chlorohydrocarbons and sterols. In addition to these compounds, the following groups were most frequently identified: aliphatic and aromatic hydrocarbons, phenols, hydroxy- and keto-aliphates and aromates, benzothiazoles and other sulphur and nitrogen containing compounds, organophosphates and a limited number of herbicides. Moreover, in excess of 150 organic compounds were detected in the sediment and suspended solids samples.

To continue with the success of the first survey, the second Joint Danube Survey was conducted in August and September 2007. During the survey 96 sites were sampled along the 2500 km of the Danube. Moreover, parallel surveys on 10 major Danube tributaries were carried out simultaneously.

The samples collected during the second survey included water, sediment, suspended particulate material, macrozoobenthos, phytobenthos, phytoplankton, macrophytes, fish and mussels. A smaller minority of the analytical work was performed on board and included physico-chemical analyses, microbiology and radon measurement. Laboratory analyses of chemical parameters included heavy metals and organic compounds primarily from the EU list of priority substances, but several other groups of compounds, such as pharmaceuticals were covered as well. A special programme of radioisotope analysis was focused on contamination by radionuclides and also on the application of isotopes for nitrate tracking and for better understanding of hydrological and biogeochemical cycles. A battery of ecotoxicological tests was part of the survey programme to obtain a better understanding of the negative impacts of hazardous substances on the functioning of the water ecosystem.

All biological quality elements needed for characterisation of the ecological status of water bodies as required by EU WFD were analysed both on-board and in the laboratories after the survey. In support of the assessment of the ecological status the first ever continuous hydromorphological characterisation of the Danube was executed. It included evaluations of the general platform and sinuosity, longitudinal and lateral continuum disruptions and the main river engineering structures.

At present, the samples collected during the survey are being analysed in the JDS2 laboratories – all being centres of excellence for particular water quality parameters. The European Commission's Joint Research Centre in Ispra, Italy is carrying out wide spectrum analyses for priority substances. Other laboratories providing chemical analyses include Bayerisches Landesamt für Umwelt in Munich, TZW in Karlsruhe (Water Technology Center), Water Research Institutes in Prague and Brno, VITUKI Budapest, Apele Romane (Romanian Waters) and ICIM (National Research and Development Institute for Environment Protection) in Bucharest, Water Research Institute in Bratislava, Umweltbundesamt in Vienna and Hydrometeorological Institute in Kiev. The final report is planned for summer 2008. More impressions from the second Joint Danube Survey including some of the results of on-board analyses can be viewed on **http://www.icpdr.org/jds/**

NORMAN – Contract N° 018486 – is a Co-ordination Action funded by the European Commission, DG Research, under the FP6 – Priority 6.3 " Global Change and Ecosystems "

NanoInteract: Development of a platform and toolkit for understanding interactions between nanoparticles and the living world

Co-ordinator Prof. Kenneth DAWSON
University College Dublin, Ireland. kenneth@fiachra.ucd.ie
http://www.nanointeract.net/

NanoInteract is an FP6 StReP project funded under the NMP theme [1], running from January 1st 2007 until 31st December 2009. It involves nine European academic institutes, two National Research Centres, five industry partners, and one US academic institute. It is one of the larger co-operative laboratory-based projects in the arena of nanoparticle hazard funded under FP6.

In designing the project, we were aware that there are significant limitations in the direct application of the current approaches to the assessment of chemical hazard (as applied to pesticides and air pollutants, for example). The are many new issues, such as the need for controlled and reproducible dispersions of nanoparticles in biological fluids, a difficult and challenging aspect of the field. Also, we felt that there could be more subtle biological effects, as in the case of asbestos-induced mesothelioma (which still cannot be predicted by any existing toxicology test); we therefore sought to investigate the options beyond direct cytotoxicity or genotoxicity, and introduced modern approaches for the detection of biological impacts.

Above all, in attempting to develop a scientifically rational approach to understanding interactions of engineered nanoparticles with living systems, we recognised the need to include leading European experts in a wide variety of fields. The project is very interdisciplinary, which is probably the only way that true excellence in this arena can be built at present, no one discipline having sufficient skill to carry out such a programme. We should emphasize that building such a capacity is an enormous and challenging task in many arenas of science, and significant barriers are met and dealt with as they arise in the programme. Given the magnitude of the task, and the importance of the topic, NanoInteract has remained in contact with many scientists across Europe and other countries, to exchange experiences to date. At the recent ESF-EMBO [2] meeting in Sant Feliu de Guixols, experts form all over the world discussed the issues, and NanoInteract scientists were highly active in that meeting.

NanoInteract seeks to identify the routes via which nanoparticles enter and accumulate in cells. Using advanced methods of chemical, physical, biological and toxicological sciences, we then connect nanoparticle properties (in physiological conditions) to the mechanisms via which they interact with, and disrupt, cellular processes. Fundamental to the project is the effort to establish protocols and standards for controlling every step of the project as we seek to eliminate the factors that currently cause irreproducibility. An overview document of these protocols will be published as an output of the project to enhance progress in the field and to share our experiences in this arena with others.

The NanoInteract project combines state-of-the-art techniques, methodologies, skills and instrumentation from all of the relevant scientific arenas to make discipline-independent platforms to address key questions of particle quality, dispersion, imaging, and biological functional impacts. Significant industrial representation is included. It is divided into six inter-connected workpackages. Ecotoxicology informs the research arena of the expected levels and type of exposure in the coming decades; toxicology gives lead information on which of these systems should be looked at in more detail. Particle science, physical, biophysical chemistry, and instrumental science ensure that the systems are characterised to a level not previously achieved, thereby enabling a rational connection to the biological processes. Biophysics and cell biology combine with molecular biology to identify the location of the nanoparticles on the cell surface, or inside the cell. These then combine with modern biological technologies and health science to connect the insult induced by the nanoparticle system to biological processes and potential disease classes that could emerge as a consequence.

One interesting and important hypothesis that is gaining some support from the work is that the surface of nanoparticles may vary significantly, even if the nanoparticles seem identical using conventional techniques. Furthermore, it has been proposed that the biological impacts are related to attachment of proteins (and other biomolecules) to this surface, and the resulting 'corona' of biomolecules has been studied (see Figure 1). The nature of the attached biomolecules has been identified and there is some hope that it will be possible to classify nanoparticles based on this approach. Similar ideas are being investigated by the ecotoxicology groups.

Overall, the experience of NanoInteract leads us to emphasise the challenging nature of this problem, requiring highly focused and large multidisciplinary teams that can address all the issues. Significant results are emerging, but there remains much more to be done.

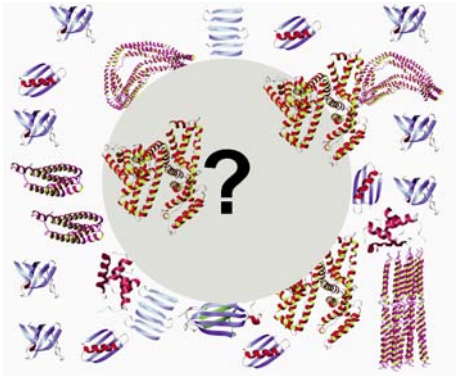


Figure 1: Plasma proteins adsorb to nanoparticles in biological fluids to form a dynamic biomolecule corona. The adsorption appears to be very selective, with several less abundant proteins being preferentially adsorbed over more abundant ones such as human serum albumin.

1 Nanotechnology and nanosciences, knowledge-based multifunctional materials and new production processes and devices
2 European Science Foundation / European Molecular Biology Organization

REFERENCES

Cedervall T, Lynch I, Lindman S et al. Understanding the nanoparticle protein corona using methods to quantify exchange rates and affinities of proteins for nanoparticles, PNAS, 2007, 104, 2050-2055.

Cedervall T, Lynch I, Foy M et al. Detailed Identification of Plasma Proteins Adsorbed on Copolymer Nanoparticles, Angew. Chem. Int. Ed. 2007, 46, 5754-5756.

NORMAN – Contract N° 018486 – is a Co-ordination Action funded by the European Commission, DG Research, under the FP6 – Priority 6.3 " Global Change and Ecosystems "

Pesticides monitoring in the Netherlands

Co-ordinators Martina VIJVER and Geert DE SNOO
Leiden University, Institute of Environmental Sciences, P.O. Box 9518, 2300 RA, Leiden, The Netherlands
http://www.pesticidesatlas.nl

1. INTRODUCTION

In most European countries, national and regional water authorities regularly monitor pesticide concentrations in surface waters. This is true of the Netherlands, but the results there are amalgamated and reported periodically by different stakeholders. The lack of transparency of monitoring data has made it difficult to make optimal use of these data in, for example, evaluating environmental policy goals, re-approval of active ingredients, prioritising problem areas and clean-up actions at national level. Against this background, at the initiative of Leiden University's Institute of Environmental Sciences (CML), and with solid support from an array of other organisations, a study was conducted to assess the potential for presenting such maps in the form of a Pesticides Atlas of the Netherlands on the internet. The Pesticides Atlas aims to provide an insight into pesticide contamination of Dutch surface waters and to investigate:

1. where a pesticide is being measured, observed and exceeding an environmental standard;
2. how to improve the regional monitoring systems;
3. whether it is possible to link pesticide concentrations with land use data (related to the re-approval of pesticides, identification of problem pesticides and areas);
4. whether pesticides concentrations change over time.

The core business of the internet tool is information at the level of separate active ingredients. Today, Dutch pesticides monitoring data have now been processed into a freely available graphic format accessible on-line (www.pesticidesatlas.nl).

This contribution will focus on the first two aims.

2. MATERIAL AND METHODS

2.1 Processing and aggregation of monitoring data
Pesticide monitoring data for the periods 1997-2006 were derived from databases owned and administered by 28 Water Boards, with prior checks being made on data quality and quantity. Environmental quality standards used to review pesticide concentrations are the European drinking water standard, the Maximum Tolerable Risk (MTR) and the pesticide authorisation standard applied by the Dutch Board for the Authorisation of Plant Protection Products and Biocides (CTGB). The drinking water standard has been set at 0.1 µg/l for almost every individual pesticide, while MTR and CTGB criteria are pesticide-specific and vary with the toxicity and the environmental, chemical and physical properties of the pesticide.

To create the pesticide maps the raw monitoring data are first processed, aggregating them in a stepwise procedure. Spatial aggregation is carried out at the level of either: 1 x 1 kilometre grid cells, or 5 x 5 kilometre grid cells. Temporal aggregation is carried out firstly over annual periods, followed by 2-years periods.

2.2 Mapping monitoring data
The percentage of pesticides exceeding environmental quality standards has been calculated by dividing, for each 5x 5 km grid cell, the number of substances exceeding the standard by the number of substances assessed. A substance is regarded as exceeding the standard if the 90th percentile of the measurements exceeds the standard. If fewer than 15 substances have been assessed, the calculation of the percentage is regarded as unreliable, and the 5x5 km grid cell is shown in grey on the maps. If no measurements are available, the cell is shown in white. Colours are used to depict the amount of exceeding pesticides. Maps are also available on the internet in which the magnitude of exceedance of measured concentrations is given. In those cases, colours are used to depict the magnitude of exceedance using classes such as 1 x MTR, 2 x MTR and 5 x MTR exceedance (data here not shown).

3. RESULTS AND DISCUSSION

Today all pesticide monitoring data for the period 1997 to 2004 have been gathered and analysed. The data have been presented on the Internet for every 2-year period (www.pesticidesatlas.nl). The number of active ingredients and the number of locations where measurements have been carried out are shown in Table 1. From this table it is clear that, for every period, data are available for about 200 active ingredients. The number of locations has increased over time. For every 2-year period there are about 150,000 measurements available, but within that total, the numbers for each active ingredient vary greatly, as does the geographical distribution of pesticides measured across the Netherlands.

Table 1. Overview of the quality of the pesticide data in the database.

2-year period	No. of active ingredients	No. of locations
1997/1998	199	512
1999/2000	187	717
2001/2002	216	781
2003/2004	291	877
2005/2006 *	430	900

* preliminary monitoring results, data available in the Pesticides Atlas in December 2007

3.1 Pesticide monitoring data and environmental standards
In the Pesticides Atlas information is presented of all pesticides combined. Figure 1 illustrates that in the Netherlands the European drinking water standard (0.1µg/l) is being exceeded in at least 30% of the locations in all months of the year. It is remarkable that pesticide concentrations were also above this standard during the winter period, because hardly any pesticides are used on most crops in this period. The percentage of locations where the pesticide authorisation standard is being exceeded is much lower (max. 14%, Figure 1). This can be explained by the different values of the quality target and the fact that not all pesticides that exceed the EU-DWS are legally allowed.

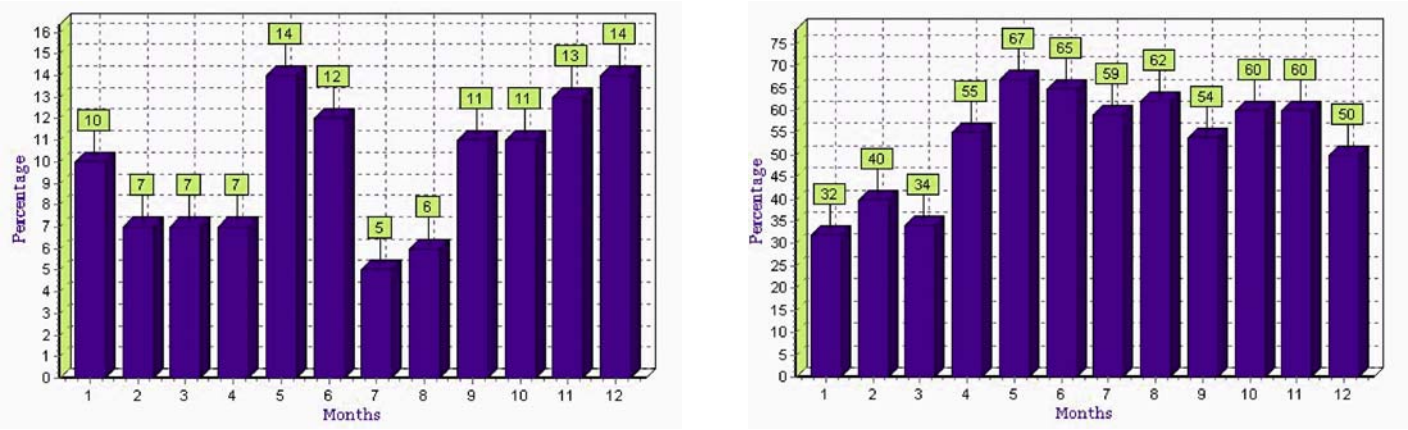


Figure 1. Percentage of locations exceeding an environmental standard during the year (2003/2004): left side European drinking water standard (0.1 µg/l), right side authorisation standard.

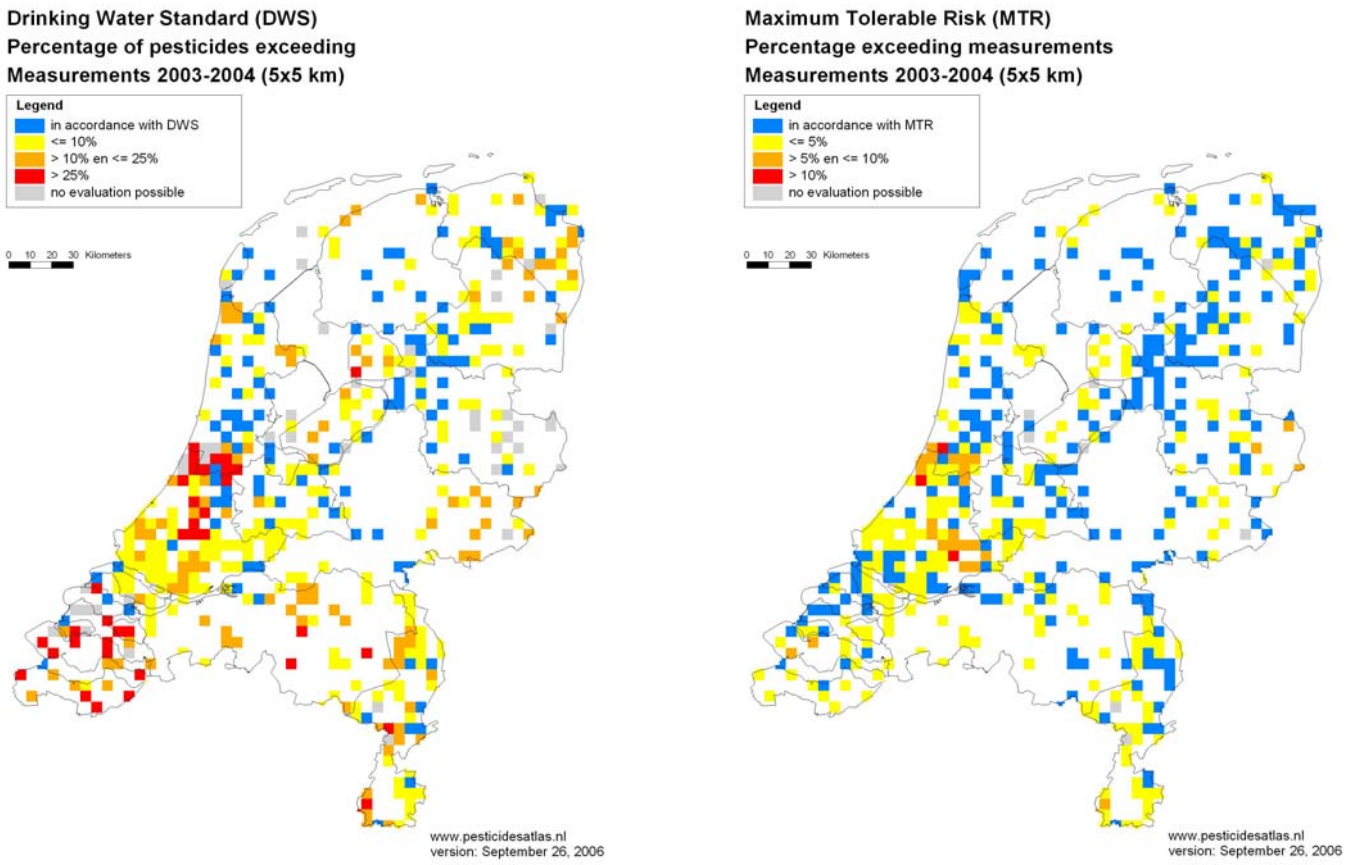


Figure 2. illustrates the percentage of pesticides in the Netherlands that exceed the European drinking water standard (0.1µg/l) and MTR over the periods 2003-2004. Many sites can be found that have at least 10% of the pesticides measured exceeding a target value. Regional differences are evident in these figures. In the western part of the country, in particular, many pesticide concentrations exceed the standards. Left side: Percentage of pesticides exceeding the EU drinking water standard during the period 2003-2004. Right side: Percentage of pesticides exceeding the MTR during the period 2003-2004. These maps are plotted on a 5 x 5 km grid scale.

3.3 Improvement of monitoring programmes

One of the aims of the Pesticide Atlas is to assist in building an optimal monitoring programme. The costs of gathering data on pesticide concentrations in the various surface waters are very high, e.g. costs of sampling, analysis and data interpretation. Yet many Dutch Water Boards could improve their programmes by investigating which pesticides are allowed to be used in their areas and watercourses that feed in to their districts. The necessity of this is illustrated in Figure 3, where measurement data on Metribuzin for the period 1999-2000 are shown (left side), together with (right side) a map of land use data – here, potato fields. The pesticide Metribuzin is used almost exclusively on potatoes. It is obvious that the monitoring in the south-east part of the country is questionable. In contrast, in the polders in the middle of the country, many potato fields can be found, but there are no measurements of this pesticide which is widely used on these crops (see the arrows in the Figure 3).

NORMAN – Contract N° 018486 – is a Co-ordination Action funded by the European Commission, DG Research, under the FP6 – Priority 6.3 " Global Change and Ecosystems "

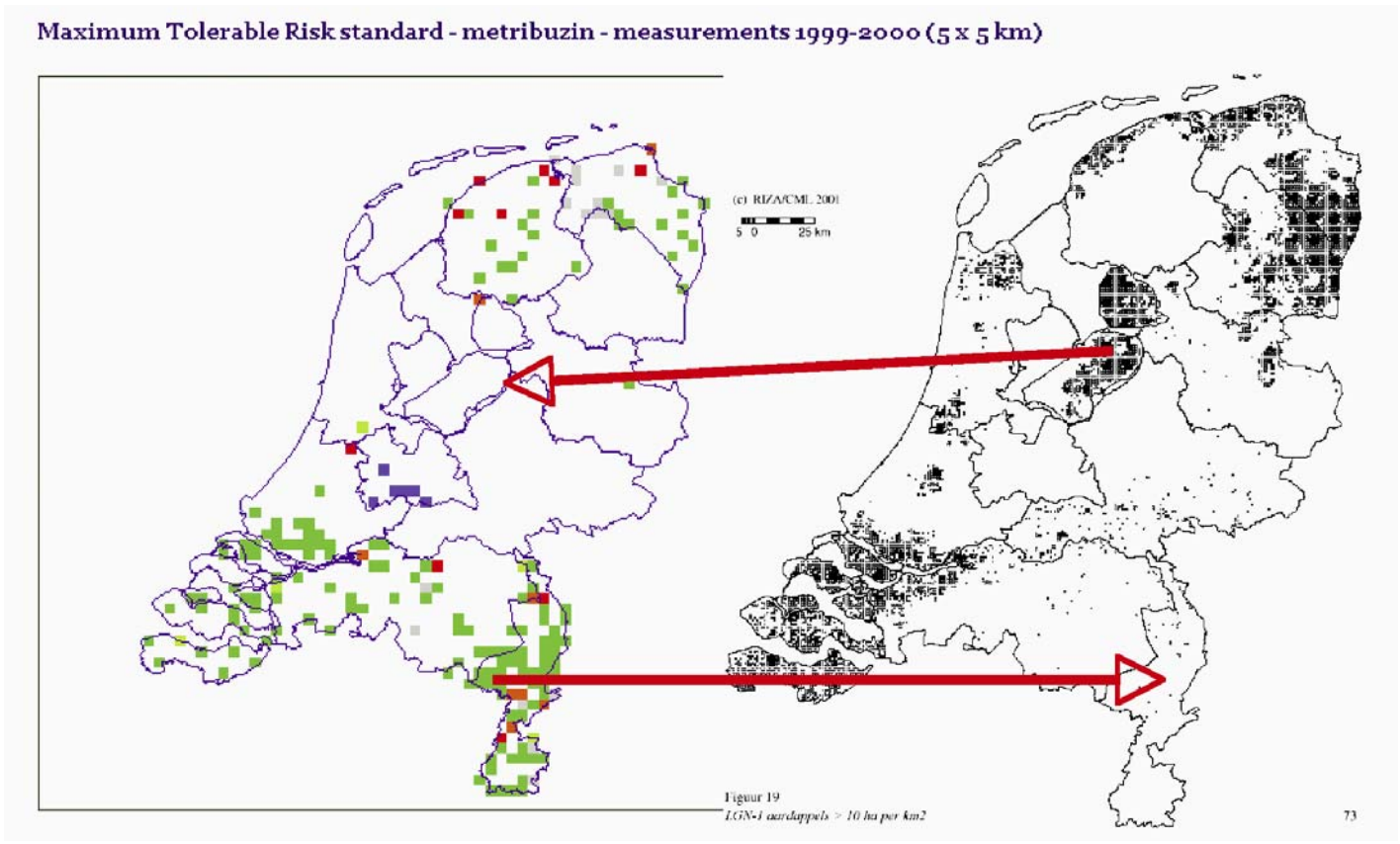


Figure 3. Example of measurements performed on Metribuzin, a pesticide allowed to be used only in potato growing. On the left side: measurements of Metribuzin. On the right side: potato fields in the Netherlands.

In the Atlas, statistical correlations have been computed between surface water pesticide concentrations on the one hand and the areas devoted to types of land use (generally types of crop) on the other. Statistical relationships that are scientifically sound could be derived for 249 active ingredients - land use combinations over the four monitoring periods. These relationships can be used, for instance, to optimise the monitoring strategies.

Results show that at most locations in the Netherlands a low percentage of the expected pesticides is measured. Actually, nowhere does this exceed 75% and the percentage of measured pesticides varies greatly between regions. In general, monitoring programmes could be substantially improved across the whole country by targeting pesticide measurements on the pesticides that local land use would lead one to expect to find.

4. CONCLUSIONS

The unique way of mapping pesticide concentrations adopted by the Pesticides Atlas enables transparent communication to a broad user-group such as national and local water managers. After all, the spatially explicit representation of data has several advantages compared with traditional (tabular) presentation. With the actual tool on the internet, a wide range of actors such as policy makers, regulators, farmers, the chemical industry, the food industry and NGOs can see a good visual representation of the geographic spread of pesticide concentrations in surface water in the Netherlands. For more information have a look on **www.pesticidesatlas.nl**

EU Neptune Project, new Sustainable Concepts and Processes for Optimisation and Upgrading of Municipal Wastewater and Sludge Treatment

Co-ordinator Prof. Dr. Hansruedi SIEGRIST
Eawag, The Swiss Federal Institute of Aquatic Science and Technology, Switzerland
http://www.eu-neptune.org

The limited supply of natural resources, global demographic trends and new legislation, have brought a change in the concept of wastewater treatment; instead of being an end-of-pipe solution for the reduction of nutrients and pathogens in the effluent before discharge, waste water treatment plants (WWTP) are delivering resources for the environment and for human activities.

The major objective of NEPTUNE is to implement sustainable technical solutions into WWTP so that new goals are better achieved. This will be accomplished either by upgrading existing municipal infrastructure or by developing new techniques.

Project NEPTUNE is financially supported by the EU as a specific targeted research or innovation project under the Sixth Framework Programme of the European Commission. Experts from 18 partner institutions from Europe, Canada and Australia, divided into 6 work packages (Fig. 1), aim to pool - and further develop – their knowledge so as to:

- « control and evaluate the (eco-)toxicity of treatment plant effluents and the discharge of organic pollutants;
- improve nutrient removal and recycling;
- optimise energy consumption and produce energy out from organic pollutants;
- ensure safe and sustainable sludge handling, reuse, inertisation and disposal;
- enable better comparability of various treatment options ».

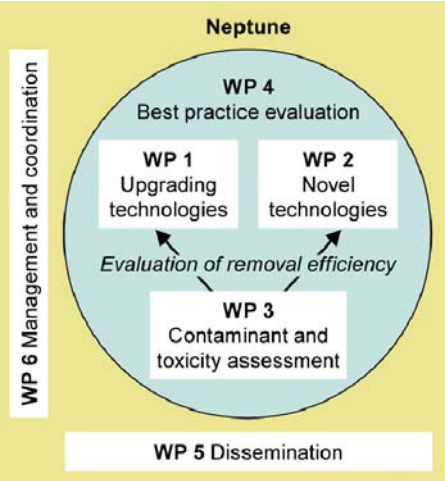


Figure 1: Work packages within NEPTUNE

CONTAMINANT AND TOXICITY IDENTIFICATION AND ASSESSMENT (WP3)

Control of chemical and microbial water quality is a crucial aspect to assess the appropriateness of mitigation technologies for municipal wastewater. The number of organic pollutants detected in surface waters has increased dramatically during the last two decades [1, 2]. Most of these emerging pollutants originate from municipal wastewater. This is especially the case for ingredients of products which are consumed by humans (e.g. food, pharmaceuticals) or just used in households (e.g. personal care products, washing agents, disinfectants, flame retardants used in electronic devices). The fate (sorption, degradation, mineralisation) of organic pollutants is often poorly understood. Even a reduction in toxicity cannot be taken for granted because compounds with comparable or even higher biological activity may be formed during advanced treatment. Such transformation products can be identified by sophisticated analytical techniques, while their biological activity and toxicity can only be evaluated using biotests. In this context, TIE (Toxicity Identification Evaluation) will be used to identify compounds of concern that may arise during advanced treatment.

A major objective of Neptune is therefore to combine sensitive chemical analyses for micropollutants and biological toxicity testing of whole effluents (Fig. 2,Tier 1, 2 testing)

Most compounds already listed as priority substances (WFD) and POPs can be covered by the consortium (e.g. alkyl phenols, alkylphenolethoxylates, polybrominated diphenyl ethers, tin organics, PAHs, organochlorines, pesticides and related metabolites). In addition to POPs and WFD priority substances, the emission of so-called 'emerging' chemicals is of major environmental and drinking water relevance. Due to the large number of emerging compounds (e.g. pharmaceuticals, perfluorinated compounds, biocides) only a small subset can be analysed in NEPTUNE. The final selection of the target compounds for NEPTUNE will be based on distinctive physical-chemical properties of the analytes, to facilitate the extrapolation to pollutants with similar properties.

Selection criteria are:

- « Strongly sorbing contaminants with high-to-medium persistence, non specific or specific sorption;
- Highly dissolved compounds with high-to-medium persistence;
- Dissolved and sorbing compounds with high-to-medium persistence ».

In addition to more innovative emerging pollutants, organic tracer compounds the behaviour of which is already known (e.g. pharmaceuticals) will also be included in the analytical methods, to facilitate the extrapolation of the results to other emerging groups.

Elucidation of biodegradation products in biologal treatment under different conditions will be another major task of Neptune (WP 3). For individual compounds, the chemical structure of stable transformation products will be identified to elucidate the degradation pathways under typical biological wastewater treatment conditions. The ecotoxicity of the emerging compounds and transformation products will be roughly determined using the Tier 1 testing approach outlined below.

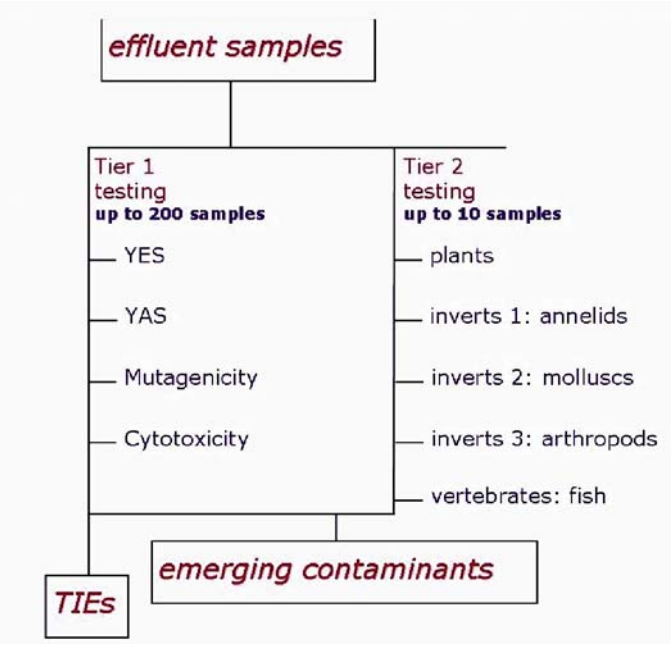


Figure 2: Overview of testing programmes for toxicity testing at Tier 1 and 2 within NEPTUNE

REFERENCES

[1] Petrovic, M., Gonzalez, S., Barcelo', D. (2003) *Analysis and removal of emerging contaminants in wastewater and drinking water*, Trends Anal. Chem., 22: 685-695.

[2] Richardson S.D, Ternes T.A. (2005) *Water Analysis: Emerging Contaminants and Current Issues*, Anal. Chem., 77: 3807-3838.

NORMAN – Contract N° 018486 – is a Co-ordination Action funded by the European Commission, DG Research, under the FP6 – Priority 6.3 " Global Change and Ecosystems "

Life of the network, NORMAN Activities

*The activities of the NORMAN project started officially on 1st September 2005, with a kick-off meeting in Paris on 7-8 September 2005. We are laying the foundations on which to build the services that will be provided by the network. The ultimate aim is to meet users' needs in the exchange and production of good-quality and comparable data in a field where data are typically scarce and insufficient for sound decision-making. Below is a summary of the activities carried out so far and forthcoming results. More information on each of these activities is provided on the project website (**www.norman-network.net**).*

NORMAN CONTACT POINTS

The network of NORMAN National Contact Points continues to grow, with the following additions to the list during the second part of 2007:

- **Portugal** – Maria de Fatima Alpendurada, of IAREN - Water Institute of the Northern Region;
- **Romania** – Zaharie Moldovan, of the National Institute of Research and Development for Isotopic and Molecular Technology;
- **Wallonia region of Belgium** – Philippe Maetz, of ISSeP - Institut Scientifique de Service Public;
- **Cyprus** – Maro Christodoulidou, of the State General Laboratory;
- **Moldova** – Anna Cumanova, of the State Hydrometeorological Service;
- **Serbia** - Zoran Stojanovic of RHMSS.

We are grateful to all our National Contact Points for taking on this vital role in furthering the exchange of information on emerging substances.

WORKSHOPS

Workshop on New tools for bio-monitoring of emerging pollutants, Amsterdam, the Netherlands: 29–30 October 2007

IVM Vrije Universiteit organised the successful second thematic NORMAN workshop, the focus of which was:

- the state of the art and future developments in the field of bio-monitoring;
- an overview of the tools currently available, from the laboratory to the field, and;
- the approaches to the identification of emerging pollutants, such as the EDA / TIE protocols.

The book of abstracts and presentations of the speakers are already available on the NORMAN site at the page **http://www.norman-network.net** ⇢ Workshops. The workshop report will be published by January 2008 on the same website.

One more workshop will be organised in Lyon, France - Spring 2008 - on **'Integrated chemical and bio-monitoring strategies for risk assessment of emerging substances'**. This workshop will focus on:

- the strategies already implemented which integrate biological and chemical methods for the identification and risk assessment of emerging substances, and;
- the gaps in these strategies regarding, in particular, the need for harmonisation of the protocols.

NORMAN DATABASES

EMPOMAP: The web-based database was published on the project website in February 2007. The process of populating the database continues apace, and the information is regularly updated. So far 87 projects, 45 experts and 24 organisations, operating in the field of emerging pollutants are registered in the database. This is a pretty good achievement, given that progress is generally slow at first with all such undertakings. However, quite a lot of effort will still need to be put by the NORMAN partners and Contact Points in order to encourage more active involvement in this task. We invite all experts around the world working in the field of emerging substances to register in the database their profiles as experts, and to contact INERIS (valeria.dulio@ineris.fr) for registration of relevant research in the field of emerging substances.

EMPODAT: the EMPODAT database, with the accompanying data collection tools (Electronic templates for data collection) has been successfully developed and tested by the partners. The database is currently being populated with the datasets from the NORMAN partners and newly registered members of the NORMAN network. The database has been available since February 2007 on the Environmental Institute (Slovakia) website, accessible to all NORMAN partners and potential data providers who have registered as members in EMPOMAP. However, the database will be published on the public website only in the final phase of the project (May 2008) due to the time needed for inclusion of the technical comments by the data providers and achievement of a critical mass of data entries.

EMPOMASS: the EMPOMASS database has also been available since February 2007 on the Environmental Institute website with access restricted to partners of the consortium and external organisations that have registered as members of NORMAN in the EMPOMAP database. A first set of data (⇢7000 entries) has been uploaded for testing. As with EMPODAT, the database will be published on the public website only in the final phase of the project (May 2008).

QA/QC ACTIVITIES

Methods validation protocols

After publication of the first draft of the protocol for validation of methods for monitoring and bio-monitoring of emerging pollutants (actually three protocols, each one addressing a different stage of the validation process, including a protocol for the integration of biological and chemical test methods) on the NORMAN website in February 2007 (http://www.norman-network.net), improvement of the protocols will be made during the third year (2008), based on the comments received and on the feedback from the three interlaboratory studies organised during the project. In 2008 the network will prepare the ground for implementation in the field of European Legislation and Standardisation (e.g. at the CEN level).

Case studies

Case 1 - Oestrogens in sewage treatment effluents – The evaluation of the results of the intralaboratory study was carried out by UKEA and IVM, and a workshop was organised on the 23 October in Bergen, Norway to discuss the results with the participating laboratories. The final report will be published on the NORMAN website (expected March 2008).

Case 2 - Non-steroidal anti-inflammatory drugs – The 1st interlaboratory study was performed from October to December 2006. The meeting of the participating laboratories was organised by JSI in Ljubliana, Slovenia on 16 April 2007 and the report is available on the NORMAN website. The second round of the interlaboratory study is under way. The deadline for sending the results is 31 October and the statistical evaluation is planned for February 2008.

Case 3 - Brominated flame retardants (DecaBDE) - the 1st interlaboratory study took place between September and November 2006. A standard solution and a dust sample were provided. A written report on results of the 1st interlaboratory study was published in June 2007 on the NORMAN website. The laboratories for the second round of the Case 3 interlaboratory study have been selected and the test materials (sediment and dust sample) prepared. The 1st meeting of the participants in the C3-II interlaboratory study will be organised in January 2008. The second round of the interlaboratory study will be performed from December 2007 to January 2008. This round will focus on the transfer of knowledge on decaBDE analyses to the routine laboratories, and on testing the V3 protocol.

Forthcoming events

LIQUID CHROMATOGRAPHY-TANDEM MASS SPECTROMETRY FOR SCREENING AND TRACE LEVEL QUANTITATION IN ENVIRONMENTAL AND FOOD SAMPLES

7 ⇢ 8 February 2008, Barcelona, Spain
4th International Workshop organised by Consejo Superior de Investigaciones Científicas, Barcelona, Spain and Institut d'Estudis Catalans, Barcelona, Spain
For more details: **http://www.cid.csic.es/barcelona2008/objectives.htm**

INTERNATIONAL CONFERENCE ON PHARMACEUTICALS PRODUCTS IN THE ENVIRONMENT: TRENDS TOWARDS LOWERING OCCURRENCE AND IMPACT

19 ⇢ 20 February 2008, Nîmes, France
Organised by the Ecole des Mines d'Alès, ARMINES, the European Federation of Pharmaceutical Industries and Associations (EPFIA) and the Society of Chemical Industry (SCI) as part of the EU-funded project KNAPPE
For more details: **http://www.knappe-eu.org/**

NORMAN IS PREPARING THE FOLLOW-UP

CALL FOR EXPRESSION OF INTEREST

Are you involved in research on the occurrence and environmental effects of emerging substances? Or are you concerned with the assessment and management of the risks associated with them?

If so, you are a potential stakeholder in the NORMAN network, the successor to the NORMAN Project. Starting from September 2008 this new, non-profit association will be a permanent network open to you and all interested stakeholders dealing with emerging substances.

What will be the benefits for your organisation as a member of the future NORMAN permanent network?

- Active involvement at an early stage in the debate on highly strategic topics. Today's emerging substances will probably be among tomorrow's regulated substances;
- Fast and easy access to the available information on institutes, projects and experts in the field of emerging substances in Europe and beyond:
  - ⇢ opportunities for collaborative research with other research institutes;
  - ⇢ more effective support to public authorities;
- A bigger voice speaking to the EC and other public institutions:
  - ⇢ the EC wishes to address NORMAN as an independent and competent forum for an overview of the state of the art for topics of concern, research needs and priorities for future regulations;
  - ⇢ visibility and credibility in the field of emerging substances;
- Adding value to activities and initiatives at the national level.

Who should be part of the network?
All interested stakeholders dealing with emerging substances – whether in studying their occurrence and effects or risk assessment and risk management. Among the key members there will be:

- Competent authorities / Reference laboratories: i.e. institutes/ organisations designated by the competent authorities at the national level to offer technical and scientific support in specific fields related to environmental protection;
- Research centres and academia;
- Industry stakeholders;
- Government institutions and standardisation bodies.

You will find more details about:

- what you will get from this permanent network;
- the advantages to your organisation of being involved in it;
- the membership fees;

on the new welcome page of the NORMAN website (**http://www.norman-network.net**).

If you think that your organisation will wish to become member, please contact:
Valeria Dulio
Scientific Co-ordinator of the NORMAN project
INERIS, Direction Scientifique
Parc Technologique ALATA BP N°2
F-60550 Verneuil-en-Halatte
E-mail address: valeria.dulio@ineris.fr

NORMAN is funded by the European Commission, DG Research, under the Sixth Framework Programme – Priority 6.3 "Global Change and Ecosystems". Contract N° 018486. This newsletter does not represent the opinion of the EU Commission. The EU Commission is not responsible for any use that might be made of the data appearing within this newsletter.

>>>>>>>>>>>>>> *Contributing to the Newsletter*

If you would like to contribute to a future issue of the newsletter or announce an upcoming event, you can contact the newsletter co-ordinator at corinne.mandin@ineris.fr
The next issue of the newsletter will be published in April 2008.

NORMAN – Contract N° 018486 – is a Co-ordination Action funded by the European Commission, DG Research, under the FP6 – Priority 6.3 " Global Change and Ecosystems "