



RECETOX NEWSLETTER

The RECETOX NEWSLETTER is a quarterly newsletter by the Research Centre for Toxic Compounds in the Environment (RECETOX), Brno, Czech Republic.

Learn,
discover,
prove
and apply



RECETOX is an independent REsearch Centre for TOXic Compounds in the Environment operating within the Faculty of Science, Masaryk University, Brno, Czech Republic. The Centre fulfills three roles: an academic institution providing university education, a research institution working on transformation of research into practical applications and a body supporting implementation of the Stockholm Convention on Persistent Organic Pollutants (POPs) and of sound chemicals management in general, nationally and internationally.

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- ▶ Echoes of the 11th Summer School on Toxic Compounds
- ▶ Sharing the RECETOX Research Infrastructure
- ▶ BIOGATE SoMoPro project



Editorial

Dear readers,

We hope you had a good time in the summer and during your holidays. We at RECETOX Centre have continued our work and implemented activities during the summer as well as strengthened our team. Among others, we finalized a set of four training videos for UNEP on procedures necessary to have clean sampling media for passive sampling of ambient air for monitoring of persistent organic pollution. In addition, several new international staff for RECETOX were recruited abroad and new student and researcher mobility projects are available through projects supported by the EEA grants.

Moreover, we continued to share our expertise at a special event organized for LIPKA (ecological NGO supporting awareness-raising), at the special OpenDay for secondary schools, as well as with the general public at the Night of Scientists. Let`s look back together at the events and meetings held at RECETOX between June and the end of September from the 11th International Summer School with a focus on cyanobacterial toxins and their effects as well as building capacities in support of the implementation of the Stockholm Convention to the 3R Alternatives to Toxicological and Pharmacological Testing Workshop in September.

We wish you pleasant reading, a successful start of the academic year to RECETOX students and teachers, and a fruitful autumn.

Katka Šebková
on behalf of October issue editors

PS – The RECETOX newsletter is also available automatically if registered through www.recetox.muni.cz or newsletter@recetox.muni.cz and it is published in English and Czech. The next issue will be released in December 2015.



Calendar of Events

- ▶ 23–28 August 2015 **Dioxin 2015 International Conference**, São Paulo, Brazil
- ▶ 30 August–3 September 2015 **27th Conference of the International Society for Environmental Epidemiology - Addressing Environmental Health Inequalities**, São Paulo, Brazil
- ▶ 17 September 2015 **OpenDay, RECETOX**, Brno, Czech Republic
- ▶ 24 September 2015 **3R Alternatives to Toxicological and Pharmacological Testing Workshop, RECETOX**, Brno, Czech Republic
- ▶ 25 September 2015 **Night of the Scientists**, Brno, Czech Republic
- ▶ 25–26 September 2015 **Advisory Group on Endocrine Disrupting Chemicals**, Geneva, Switzerland
- ▶ 27 September–2 October 2015 **4th Meeting of International Conference on Chemicals Management (ICCM4)**, Geneva, Switzerland
- ▶ 29 September–2 October 2015 **BAT-BEP Expert Meeting, Stockholm Convention**, Bratislava Slovakia
- ▶ 5–8 October 2015 **Annual directors meeting of the Stockholm and Basel Conventions Regional Centres**, Geneva, Switzerland
- ▶ 5–8 October 2015 **Meeting of the Global Coordination Group**, Geneva, Switzerland
- ▶ 6 October 2015 **Workshop Implementation of Multilateral environmental agreements as a part of the system of chemical and environmental safety of the Russian Federation and EAEC countries**, Moscow, Russian Federation
- ▶ 15 October 2015 **20th meeting of the Council of the National Centre for Toxic Compounds**, Prague, Czech Republic
- ▶ 19–23 October 2015 **POPRC Meeting**, Rome, Italy
- ▶ 10–12 November 2015 **GEO Ministerial Conference**, Mexico City, Mexico
- ▶ 23–27 November 2015 **Training on Toxic Compounds in the Environment for laboratory technicians, Kazakhstan in RECETOX**, Brno, Czech Republic
- ▶ November 2015 **Awareness raising workshop on the Stockholm Convention**, Czech Republic
- ▶ 3–6 November 2015 **13th HCH and Pesticides Forum**, Zaragoza, Spain



Courses and Open-Access at RECETOX

11th International Summer School 2015



Visit at the Mendel Museum in Brno

The 11th International Summer School on Toxic Compounds organized jointly by the Stockholm Convention Regional Centre in the Czech Republic (RECETOX), RECETOX Research Infrastructure in cooperation with UNEP, the Ministry of Environment of the Czech Republic and CYNACOST (EU COST ES1105 Action) was held on the RECETOX premises in Brno, Czech Republic from 15-20 June 2015. This year, 42 students from 28 countries took part in the course.

Some followed the special topic of the year: “Cyanobacterial blooms and their toxins in water reservoirs,” and that brought them six intensive days filled with lectures, laboratory exercises, hands-on training, two site visits and one field trip. The Nové Mlýny reservoir site visit provided the participants with the opportunity to experience a site heavily affected by toxic cyanobacteria, sampling methods and field assessment. The collected samples were processed by the participants themselves during the follow-up laboratory analyses (bloom characterization, cyanotoxin extraction and HPLC analyses, and ecotoxicological testing) at the RECETOX premises.

In addition, a parallel class organized in cooperation with the Secretariat of the Stockholm Convention provided in depth insight and training in support of the Global Monitoring Plan under the Stockholm Convention. The two days on the GMP comprised hands-on work with sampling devices, lectures and fun competition in practical work with the online GMP tool, as well as an introduction to the laboratory facilities and instrumentation of the RECETOX premises.

Moreover, all participants also travelled to the Košetice Observatory, a sampling site used in MONET and EMEP (European Monitoring and Evaluation Programme) monitoring programmes as the Central European background site for

research on the long range transboundary air pollution and visited Mendel Museum in Brno to learn about one of forefathers of genetics at the 150th anniversary of his birth.

Lectures on cyanobacteria were provided by leading European experts in all aspects relevant to cyanobacteria, i.e. ecology of toxic cyanobacterial blooms (prof. Bas Ibelings, University of Geneva, Switzerland), health hazards (prof. Zorica Svircev, University Novi Sad, Serbia) ecotoxicological risks (prof. Luděk Bláha, RECETOX), cyanobacterial toxins and their analyses (prof. Jusi Meriluoto, Abo Academy University, most recent undertakings in research, new experiences and options for solving practical challenges and cases.

Hands-on a passive sampler deployment.





The whole week provided the opportunity for close interaction with lecturers and RECETOX staff during the courses (Turku, Finland), and remediation of degraded reservoirs (prof. Blahoslav Maršálek, (Czech Academy of Sciences and RECETOX)). Invited lecturers were complemented by a dozen of the RECETOX staff lecturers providing updates on the most recent undertakings in research, new experiences and options for solving practical challenges and cases.

The whole week provided the opportunity for close interaction with lecturers and RECETOX staff during the courses, laboratory hands-on activities, and during the breaks and field visits. This summer school received a great reviews in all areas based on feedback questionnaires completed by participants, who acknowledged high standards, practicality of the course and interconnected educational activities. This is a selection of the responses from the questionnaires or follow-up e-mail communications:

"I learned how to prepare a passive sampler. It was helpful and interesting - the lectures on GMP online tools. Now it will be easier for me to find out concentrations of POPs quickly for comparing or for studies. I am very satisfied because I visited many working lab and I realized that instruments are very sophisticated. It was very helpful for me especially because I improved my knowledge on new methods of sampling."



Košetice field trip and introduction to a range of monitoring equipment.

"It was my pleasure to be participant of this course. Thank you!"

"I want to thank you for the really good material and information that you shared with us"

"Practical training in ecotoxicology was very, very interesting and helpful, for sure!"

Dear all!

By the present, let me thank you for the perfectly organized 11th summer school in Brno. It was extremely informative and I am sure that it will be useful not only for me, but in general will help to improve implementation of Stockholm convention in Russia.

Especially, thank you for your time during the social program and all your hospitality. Hope for the future cooperation.

Best regards,

Ekaterina Ivanova

Russian Federation, UNIDO Center for International Industrial Cooperation in the Russian Federation

Dear friends,

I would like to thank you once again for your hospitality in Brno!!! For me was really pleasure to be a part of RECETOX Summer school 2015! I appreciate your big efforts to realize it! Please accept my sincere appreciation for your highly professional skills. This event has laid down a solid ground floor for the further deepening of our co-operation.

With my kind regards,

Yours sincerely,

Nina

Nina Nikolova, Institute for Nuclear Research and Nuclear Energy, Bulgarian Academy of Science, Bulgaria

Last but not least, we would like hereby to thank everyone involved in or supporting the summer school this year and we look forward to welcoming participants at the 12th course organized in June 2016.

3R Alternatives to Toxicological and Pharmacological Testing Workshop



RECETOX organized a one-day international workshop for experimental biologists, pharmacologists and toxicologists, life sciences researchers, and students at all levels of expertise in its premises in Brno, Czech Republic on 24 September 2015. The workshop entitled "3R Alternatives to Toxicological and Pharmacological Testing Workshop" provided a deeper insight into the 3R ethical principle (Replacement, Reduction and Refinement), which promotes alternative tools to minimize unnecessary suffering of laboratory animals. The workshop comprised both lectures as well as practical training in alternative in vitro models and techniques and is the first in the series of such activities that would look more closely into alternatives to conventional testing.



RECETOX Research Infrastructure

The RECETOX Research Infrastructure (RI) is a distinct part of the RECETOX Centre. The Centre studies challenges and linkages between the environment and health. Four research programs and three core units form the Centre. These three core units are managed together as the RECETOX RI, providing its users with an ever-expanding range of expertise and services through transnational open-access.

The RECETOX RI is a typical multipurpose infrastructure; its three core units carry out their own long-term projects, generating new and applying available data, while at the same time they offer a broad portfolio of laboratory and IT capacities, expertise and services. The portfolio includes the development of materials and novel techniques for sampling (10% of the users), chemical analyses of (samples of) environmental matrices for a broad spectrum of contaminants (40% of the users) and laboratory studies of environmental processes (5% of the users). The expertise available also allows for design, synthesis and characterization of new photoactivatable groups and supramolecular complexes through environmental technology (5% of users), as well as reaction mechanism studies. The RI also offers the development of biosensors for detecting environmental pollutants, IT tools for designing biocatalysts and for screening substrates and inhibitors, as well as the development of specific catalysts for the degradation of toxic wastes (5% of the users). The RI also offers tools for analyses of chemical bioaccumulation, for identifying biochemical mechanisms and markers of stress or specific studies of mechanisms of toxicity using bacteria, algae, plants, invertebrate and vertebrate embryos, and analyses of the environmental and human exposure and risk analyses (10% of users). In addition, other IT technologies (interdisciplinary databases and web portals) are developed together with comprehensive online information systems for data management, modeling and decision-making. GIS-based visualization tools, advanced/robust statistical methods, spatial distribution and time series analyses are also available (15% of users). Finally, long-term epidemiological data describing the factors affecting the development and health of children, family environment contribution to later successes and failures of young people, and the impact of political and social changes on human health and quality of life are also available to external users (10%).

A range of partners from academia as well as public and private sector use the RECETOX RI capacities through implementation of joint or contract research projects. Many partners also use the RI's laboratory capacity, and the available environmental and epidemiological data, and visualization tools.

Open access users

We have seen more than 150 external researchers (including international ones) using the RECETOX RI services through open-access in last six years. Another 70 scientists benefited from the RI's available analytical capacity and more than 300 other participants took part in the annually organized international summer school.

Almost 75% of incoming researchers are from universities, 15% from other research institutions, 8% from the regional and national authorities, 1% from the private sector and 1% from non-governmental organizations. When looking at all user groups, the universities represent 47% of users, research institutions 19%, private sector and industry 18%, national authorities and governments 15% and non-profit organizations 1%. The private sector is much more interested in the use of available expertise and laboratory capacity than in training of their staff, while the universities and research institutions exhibit the opposite trend.

In summary, 16 international researchers and one Czech university student have used the open access to RECETOX research infrastructure between January and end of August 2015. Currently, there is Ms. Gracia Rivas Ibanez from Spain, using our infrastructure and undertaking the training in acute toxicity tests on daphnia (*Daphnia magna*).

Finally, we would like to share two testimonials from the RECETOX research infrastructure users spending time with us in 2015:

"...The Training course was excellent, I am very happy to have done it. I learned a lot of essential and very practical key information and skills. It will definitely help me for my professional life. This experience exceeded my expectations, because it not only give me hands-on research experience, but allowed me to network and take advantage of RECETOX resources that will help me with my future goals.

Very Professional place to work and friendly people...

Salvatore Cotronei, Itálie, 30 March - 30 August 2015, Laboratories of the Trace Analyses, analyses of shark tissues for levels of persistent organic pollutants

...All my expectations and more have been met. I am extremely happy that I decided to do the Erasmus program here at RECETOX, I have learnt so much which is exactly what I wanted to continue my academic career. I have so many new contacts that are renowned in their respective fields; this will certainly be helpful to me in the future..."

Orla Myers, United Kingdom, 26 April - 31 July 2015, ecotoxicology, biochemical and cell mechanisms of toxicity of organic pollutants and health risk assessment.





RECETOX Projects Insight

SoMoPro Project - BIOGATE – Rational design and engineering of enzyme gates



The first newsletter of 2014 contained an article about three new research projects granted to RECETOX in a project call by the South Moravian Programme for Distinguished Researchers (SoMoPro) for projects implemented between 2014 and 2016. That article provided a brief abstract of the each project and mentioned that individual texts will be exploring them in greater detail in the future. The last but not least of the three projects, Biogate, is currently at its half-point and we have asked its investigator, our colleague Sérgio Marques, PhD, to give us more information about the project implementation and preliminary results.

Sergio explained to us that many enzymes and other biomolecules have sophisticated structural features, so-called “molecular gates”, which open and close the access of other molecules to important regions/areas. He stated he aims to find how such gating mechanisms form, and how are their building parts orchestrated together. He uses model enzymes haloalkane dehalogenases (HLDs), which are bacterial enzymes that convert halogenated organic compounds into the corresponding alcohols. The HLDs have important biotechnological applications, namely in the synthesis of optically pure compounds, in systems for degradation of anthropogenic halogenated pollutants, and in field biosensors for detection of such pollutants.

“We study some of these proteins and the dynamics of their tunnels. We try to find the relationship with their catalytic activity by using different computational tools, such as the in-house program CAVER and simulations of molecular dynamics. We found that some of the proteins with more closed tunnels are also the most active against a common environmental pollutant, 1,2,3-trichloropropane (TCP). After identification of the key parts forming the gate in these protein tunnels, our goal is

to modify the existing gates in a rational way, in order to improve the enzyme’s activity or selectivity. Simulations of the enzymes in the presence of the substrates or products can also give important clues about their operating modes.” described Marques regarding the steps and options in the research for potential modifications of the model proteins.

Preliminary results

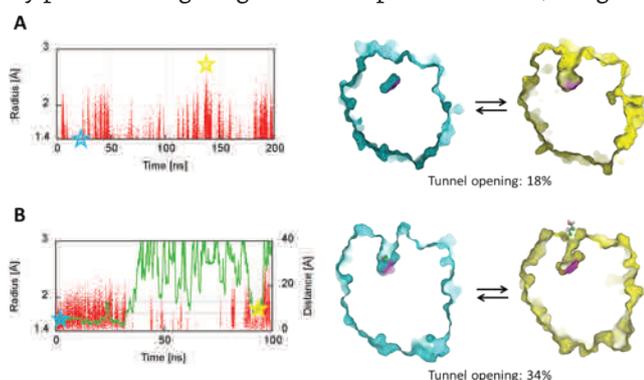
The results show that the mutant DhaA31 is currently the most efficient HLD in degrading 1,2,3-trichloropropane ($k_{cat} = 1.26 \text{ s}^{-1}$, $k_{cat}/K_m = 1050 \text{ s}^{-1}\text{M}^{-1}$). The simulations with the wild type and the mutant, with and without the alcohol and chloride products (Figure 1), have revealed the major differences in the tunnel dynamics of the two enzymes. The percentage of tunnel opening can be very different, which will have an influence on the number of solvent molecules in the catalytic site. They also bind the products and substrates with very different affinities. Altogether, these findings help us explain the catalytic differences between these proteins.

“We could also identify the key components of the gate present in the tunnel of DhaA31, such as the door, hinge and anchoring residues. These residues have been or will be targeted by mutagenesis. In this way we expect to find new mutant enzymes with more efficient gating mechanisms, and to improve their efficacy in degrading TCP or other relevant halogenated substrates”, highlights Sérgio regarding further steps.

Outlook

Sérgio concluded: “This project is now halfway to completion, and we expect that one of our main output will be the methodology for proper studying of molecular gates, and several different approaches for their engineering. By applying newly developed protocols to HLD variants, we hope to optimize existing gates and find improved mutants, which could be used in biotechnological applications in the future. At the same time, we also hope that our approach to rational design of protein gates will inspire other scientists to launch themselves into new unexplored possibilities in the field of the protein engineering.

Sérgio Marques working with software for a rational design of enzyme gates



Dynamics of the main tunnel of DhaA31 mutant during accelerated MD simulations, for the unbound protein (A) and for the protein bound with the products (B). The red dots represent the tunnel radius in each snapshot, the green line represents the distance of the alcohol product to the active site cavity, and the protein surfaces represent two of the observed conformations for each system; the catalytic residues are represented as magenta shading.



RECETOX News

Visits to RECETOX

RECETOX opened its premises and laboratories for several events in August and September. Secondary school students interested in biology could work in the RECETOX laboratories for a day in August (20.8.2015) during their week at the Rychta biological camp "Josefáč 2015" organized by Lipka. In addition, secondary school students getting ready for university studies could visit us on 17 September during the special OpenDay organized by the Masaryk University, and a yearly Night of the Scientists organized in all Europe also took place at the Bohunice Campus in Brno on 25 September 2015. We believe that all the students and families that visited us in large numbers had a great time with us.

New Team Members

We would like to warmly welcome new colleagues who joined us over the summer. Gary Codling joined our Trace Analytical Laboratories from Canada, previously-working in John Giesy's team. Gary will be working with Tono Kočan on analyses of fluorinated compounds and biological tissues. Researchers Veronika Vidová and Zdeněk Spáčil returned to Brno from the United States of America. They are joining our analytical chemists and will be sharing their experience in developing new analytical methods and capitalizing on their exper-

tise in biology. Martina Hoferková is a new member of the CELSPAC:TNG team (Central European Longitudinal Study of Pregnancy and Childhood: The Next Generation). Martina works with mothers after delivering their babies at the Brno Faculty Hospital who decide to join our epidemiological study. Pavlína Lolloková joined our project support team and among many tasks, she is in charge of keeping records of the research outcomes presented at conferences and in peer-reviewed journals. Enjoy your time at RECETOX!

International Mobility at RECETOX

We are happy to announce that our students and researchers Miroslav Brumovský, Pernilla Carlsson, and Mária Chropeňová were successful in obtaining support for projects from the Norwegian funds and EEA grants promoting the international mobility of students and researchers to Norway in coming months.

In addition, doctoral students Katarína Bányiová and Anežka Nečasová from Human Exposure Assessment and Risks research group received ISEE grant to travel to and participate at the 27th Conference of the International Society for Environmental Epidemiology (ISEE) in São Paulo, Brazil 30 August to 3 September 2015. They presented their work on Genotoxic Effects of UV filter Ethylhexyl Methoxycinnamate

in Cosmetics (Nečasová et al.) and Dermal Absorption of Polychlorinated Diphenyl Ethers (Bányiová et al.) in the poster session. Katarína acknowledged the ISEE support: "I am very grateful I had the opportunity to witness the new trends in environmental epidemiology and to discuss the results of my research with world epidemiologists. Their positive feedback is a huge motivation for my future work. In addition, the conference focussed on addressing environmental health inequalities and to see them showcased in contributions of others makes me think more broadly about environment and health issues that need to be urgently treated and that are being solved everyday in the world."

New Papers Published

RECETOX staff have so far prepared 77 research papers to be published in prestigious peer reviewed journals in 2015 and more are being finalized this year. We hereby provide a selection of new papers that came out or were accepted for publication during the summer:

- Amaro, M., Brezovsky, J., Kovacova, S., Sykora, J., Bednar, D., Nemecek, V., Liskova, V., Kurumbang, N., Beerens, K., Chaloupkova, R., Paruch, K., Hof, M., Damborsky, J., 2015: Site-specific Analysis of Protein Hydration Based on Unnatural Amino Acid Fluorescence. *Journal of the American Chemical Society* 137: 4988-4992.
- T. Ghosh, T., Slanina and B. Konig*, Visible light photocatalytic reduction of aldehydes by Rh(III)-H: a detailed mechanistic study, *Chemical Science* DOI: 10.1039/c4sc03709j
- Holla, L.I.; Linhartova, P.B.; Lucanova, S.; Kastovsky, J.; Musilova, K.; Bartosova, M.; Kukletova, M.; Kukla, L.; Dusek, L. GLUT2 and TAS1R2 Polymorphisms and Susceptibility to Dental Caries. 2015. *CARIES RESEARCH*, volume 49, issue 4. 417-424. DOI: 10.1159/00430958
- Barbora Jarošová, Jakub Javůrek, Ondřej Adamovský, Klára Hilscherová, 2015. Phytoestrogens and mycoestrogens in surface waters - Their sources, occurrence, and potential contribution to estrogenic activity. *Environment International*
- Sharma, B.M., Nizzetto, L., Bharat, G.K., Tayal, S., Melymuk, L., Sáňka, O., Přibyllová, P., Audy, O., Larssen, T., 2015. Melting Himalayan glaciers contaminated by legacy atmospheric depositions are important sources of PCBs and high-molecular-weight PAHs for the Ganges floodplain during dry periods. *Environ. Pollut.* 206, 588-596.



Upcoming Events

Conference on Air Protection in the National Administration

The RECETOX Research Infrastructure would like to invite representatives of the national, regional and local authorities as well as experts from stakeholders dealing with air contamination to participate at the 10th meeting of the national conference Air Protection in the National Administration, theory and practice taking place in the hotel Centro in Hustopeče, Czech Republic on 10-12 November 2015. The RECETOX Centre partnered with the long term organizers of the conference - Ministry of the Environment of the Czech Republic, Vodní zdroje Ekomonitor Ltd., Ascend Ltd., Bioanalytika CZ Ltd. and Institute of Chemical Technology in Prague - for the first time and brings EEA grant support.



RECETOX researchers will present outcomes of the Da Vinci project aiming at better interpretation of globally available data on atmospheric contamination by persistent organic pollutants (POPs) and more precise analysis of their longtime trends. Implementation of the Da Vinci project (Data Visualization, Interpretation and Comparison Improvements for organic pollutants in long-term monitoring networks, EHP-CZ02-OV-1-059-01-2014) is supported by EEA grants.

The conference will comprise the following expert sessions:

- European and national legislation on air protection, environmental impact assessment, integrated prevention and pollution control (IPPC) and wastes
- Mid-term (until 2020) strategy improving air quality in the Czech Republic, National Program on Reducing Emissions, programs improving air quality at the level of industrial zones and agglomerations
- Economic tools affecting environment
- Experience and lessons learned from air protection authorities and operators of air pollution sources with implementation of the Air Protection Act and its operating regulations
- Technical and non-technical measures reducing for emissions and improving air quality, climate protection synergies
- Updates on the air contamination effects and air quality monitoring

Visit www.genasis.cz and use data browser to find out more about levels of toxic chemicals around us!



The GENASIS (Global ENvironmental ASsessment Information System) created in cooperation of RECETOX with IBA MU - institutes of Masaryk University, Brno, Czech Republic - provides comprehensive information on chemical contamination of the environment, namely persistent organic pollutants (POPs). The system combines expertise, validated data from partner institutions, and input from regular environmental monitoring programmes. Users and data providers get secure data repository, sophisticated analytical tools, and comfortable data management and visualization.

Short Announcements

We are happy to announce the birth of four children in families of our young staff during the summer months 2015. Congratulations to Lenka Vaňková on the birth of her daughter Terežka, to Ondřej Adamovský for welcoming daughter Zita, to Jan Kuta on son Tomáš, and to Marie Michelová on the birth of her first child, daughter Blanka.



Trace Analysis Laboratories



Use the Accredited Laboratories and their infrastructure for the following services:

- Speciation **analyses** of toxic and essential elements in the environment and biota QA/QC system, validated analytical methods
- Modern equipment for extractions, fractionations and purification of samples
- State of the art instrumentation for analyses of endocrine disrupting chemicals, persistent organic pollutants, dioxins, furans, brominated flame retardants, steroids, explosives, and heavy metals

Long-term monitoring – air, precipitation, soil, water, sediments, needles, mosses and lichens

- Air monitoring networks – MONET in Europe, Africa and Asia
- Sampling by active and passive samplers
- Sampling of precipitation and surface waters, sediments, soils and biotic materials

Capacity building – made-to-measure training for laboratory experts on various instrumentation

Contact Dr. Petra Přibyllová (pribylova@recetox.muni.cz) for more information.



RECETOX research infrastructure provides **OPEN-ACCESS** to Czech and international researchers to work on their projects and use the expertise and instrumentation available in our Centre.

Visit www.recetox.muni.cz/RI for the application procedure.

For more information, please contact Dr. Petra Růžičková, infrastructure coordinator (ruzickova@recetox.muni.cz).



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