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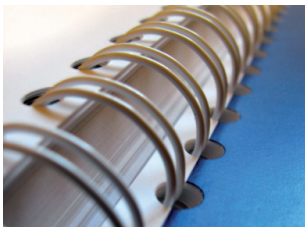


E·C·T RESEARCH AND SERVICES IN ECOTOXICOLOGY

COMPANY

ECT Oekotoxikologie GmbH is a private enterprise that was founded in 1993.

In compliance with Good Laboratory Practice (GLP) and DIN EN ISO/IEC 17025 accreditation, we perform standardised ecotoxicological tests in the laboratory as well as semi-field and field studies. Established test systems cover the aquatic, benthic and terrestrial environment. In addition to performing standardised tests, we modify existing or develop new methods to improve the characterisation of environmental risks caused by chemicals. The results of our experimental and desk-based studies are used in the environmental risk assessment of industrial chemicals (REACH), biocides, pharmaceuticals, plant protection products, wastes and genetically modified organisms.

Through constant involvement in international research projects and cooperation with universities, ECT keeps up with the state of the art in the field of ecotoxicology. Based on broad scientific expertise we can provide solutions and develop strategies to solve problems that may arise during marketing authorisation, notification or classification of chemical substances.

Since 1993, ECT has been growing constantly and employs currently about 30 permanent staff. All staff members hold a degree from a university or a technical (high) school. In addition, we host each year several undergraduate and graduate students who often conduct their thesis work with us. Our laboratory and office space covers more than 1600 m^2 and includes an isotope laboratory.

ECT is currently led by three managing directors. Jörg Römbke and Thomas Knacker († October 30, 2011) were among the founders of ECT back in 1993, while Anja Coors joined the management team in 2010. Based on their research interests and scientific experience, the three management directors cover different key activities at ECT: Jörg Römbke is specialized in terrestrial ecotoxicology as well as taxonomy and biodiversity of soil organisms, Thomas Knacker is particularly active in the field of pharmaceuticals and endocrine disruption, and Anja Coors lately focussed on mixture toxicity and multiple stressors in addition to her interest in statistics.

CONTENT

ECT conducts a broad range of ecotoxicological tests in compliance with Good Laboratory Practice (GLP) since 1994 and under ISO 17025 accreditation since 2010. We are particularly experienced in conducting laboratory, semi-field and field studies according to international standard guidelines and adapted to special requirements. If demanded by specific scientific or regulatory needs, additional custom-designed studies can be developed and performed, e.g. by applying non-standard test species, multi-species systems or modified exposure scenarios. Using non-standard test species, we are regulary establishing species sensitivity distributions.

Our testing activities (i.e. effect, fate and bioaccumulation tests) cover the main environmental compartments water, sediment and soil. The laboratory space covers more than 1100 m^2 and includes an isotope laboratory.

Analytical verification of test substance concentration, which is often required according to standard guidelines, is usually conducted by partners of ECT. Communication lines and monitoring procedures are well established with these long-term partners and ensure a smooth execution of the complete study.

AQUATIC ECOTOXICOLOGY

AQUATIC MICRO-ORGANISMS

Activated sludge, respiration inhibition test	OECD 209

AQUATIC PLANTS

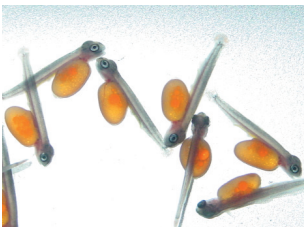
Freshwater algae and cyanobacteria, growth inhibition test	OECD 201 ISO 8692 DIN 38412-L33
Lemna sp., growth inhibition test	OECD 221 ISO 20079
Rooted aquatic macrophyte *Myriophyllum sp.*, growth inhibition test	OECD draft

AQUATIC CRUSTACEANS

Daphnia magna, acute immobilisation test	OECD 202 ISO 6341 DIN 38412-L30
Daphnia magna, reproduction test	OECD 211 ISO 10706

AQUATIC INSECTS

Chironomus sp., acute immobilisation test	OECD 235



FISH

Determination of the acute toxicity of waste water to zebrafish eggs (*Danio rerio*)	ISO 15088 DIN 38415-6
Fish, short term toxicity test on embryo and sac-fry stages	OECD 212
Fish, acute toxicity test	OECD 203
Fish, prolonged toxicity test: 14-day study	OECD 204
Fish, juvenile growth test	OECD 215
Fish, early-life stage toxicity test	OECD 210
Fish, complete life-cycle test	Draft Guideline OECD DRP (2008)

FISH (ENDOCRINE ACTIVITY)

Fish, short term reproduction assay	OECD 229
21-day fish assay: A short-term screening for oestrogenic and androgenic activity, and aromatase inhibition	OECD 230
Fish, sexual development test	OECD 234

MICRO- AND MESOCOSM TESTS

Indoor and outdoor aquatic microco - and mesocosm studies	Campbell et al. (1998) Giddings et al. (2002) SANCO/3268/2001 ASTM International E1366-02

AQUATIC INSECTS

Sediment-water chironomid toxicity test	OECD 218 OECD 219
Sediment-water chironomid life-cycle toxicity test	OECD 233

AQUATIC OLIGOCHAETES

Sediment-water *Lumbriculus* toxicity test using spiked sediment	OECD 225
Sediment toxicity test with *Tubifex tubifex*	ASTM E 1706

AQUATIC CRUSTACEANS

Hyalella azteca, sediment toxicity (28 days)	based on ASTM E 1706 and OECD 218

CHARACTERISATION OF SEDIMENT

MICROBIAL CHARACTERISATION

Determination of soil microbial biomass – Part 1: Substrate-induced respiration method	ISO 14240-1
Determination of soil microbial biomass – Part 2: Fumigation-extraction method	ISO 14240-2

MICRO-ORGANISMS

Nitrogen transformation test	OECD 216
Carbon transformation test	OECD 217
Arthrobacter globiformis — Solid contact test	ISO 10871 (draft)

TERRESTRIAL PLANTS

Seedling emergence and seedling growth test	OECD 208 ISO 11269-2
Vegetative vigour test	OECD 227
Measurement on the inhibition on root growth	ISO 11269-1
Chronic toxicity in higher plants	ISO 22030

TERRESTRIAL INVERTEBRATES

Earthworm acute toxicity test	OECD 207 ISO 11268-1 EC C.8
Earthworm reproduction test (*Eisenia fetida/andrei*)	OECD 222 ISO 11268-2
Enchytraeidae reproduction test	OECD 220 ISO 16387
Collembolan reproduction test	OECD 232 ISO 11267
Predatory mite (*Hypoaspis (Geolaelaps) aculeifer*) reproduction test	OECD 226
Non-target arthropod acute and chronic laboratory tests (surface dwellers like *Aleochara bilineata, Poecilus cupreus, Pardosa sp.*)	IOBC

MICROBIAL CHARACTERISATION OF SOIL

Determination of soil microbial biomass - Part 1: Substrate-induced respiration method	ISO 14240-1
Determination of soil microbial biomass - Part 2: Fumigation-extraction method	ISO 14240-2

DUNG ORGANISMS

Dung beetle (*Aphodius constans*) laboratory test	OECD GD 122
Dung flies (*Scathophaga stercoraria., Musca autumnalis*) laboratory tests	OECD 228
Dung organism field study	Römbke et al. (2010), Jochmann et al. (2011)

HIGHER-TIER TERRESTRIAL STUDIES

Breakdown of organic matter in litter bags in the field	OECD GD 54
Guidance on the determination of effects on earthworms in field situations	ISO 11268-3 Kula et al. (2006)
Terrestrial Model Ecosystems (TME), semi-field test for studying effects and fate of chemicals in soil	PERAS (Schaeffer et al. 2009)
Non-target arthropod semi-field tests with surface dwellers like *P. cupreus*	IOBC

TAXONOMY

ECT has expertise in the taxonomy of earthworms (Lumbricidae), potworms (Enchytraeidae), and nematodes. In several projects we have studied ecotoxicological effects on these groups of terrestrial invertebrates in combination with their ecology, distribution, and diversity in the field (monitoring studies). These projects are conducted throughout Europe as well as North and South America.

WATER

Bioconcentration: Flow-through fish test	OECD 305

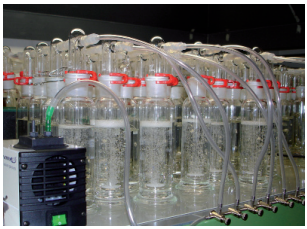
SEDIMENT

Bioaccumulation in sediment-dwelling benthic oligochaetes	OECD 315

SOIL

Bioaccumulation in terrestrial oligochaetes	OECD 317

FATE



WATER

READY BIODEGRADABILITY

DOC die-away test	OECD 301 A
CO2-evolution test	OECD 301 B
Closed bottle test	OECD 301 C
Modified OECD screening test	OECD 301 D
Manometric respirometry test	OECD 301 E
CO2-headspace test	OECD 310

INHERENT BIODEGRADABILITY

Zahn-Wellens-test	OECD 302 B

ANAEROBIC BIODEGRADABILITY

Anaerobic biodegradation in digested sludge	OECD 311

SIMULATION TESTS

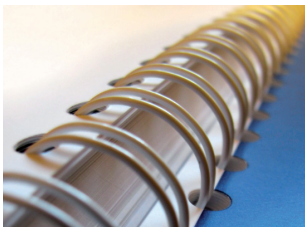
Model sewage treatment plant (coupled units test)	OECD 303 A

SEDIMENT

Aerobic and anaerobic transformation in aquatic sediment	OECD 308

SOIL

Leaching in soil columns	OECD 312
Aerobic and anaerobic transformation in soil	OECD 307



The main emphasis of ECT's consultancy is on rendering and evaluating environmental risk assessments for industrial chemicals, biocides, pesticides and pharmaceuticals. Also, we can assess the environmental risks caused by mixtures of substances and evaluate the hazardous potential of waste. Based on scientific principles we prepare expert opinions for governmental authorities as well as for chemical industry. Finally, we organise and conduct meetings and workshops where ecotoxicological topics are discussed by stakeholders.

PLANT PROTECTION PRODUCTS

Based on more than 10 years of experience ECT prepares the fate and ecotoxicology sections of dossiers for active substances and formulations of pesticides according to the latest Directives, Regulations and Working Documents released by the European Union.

By working on more than 25 pesticide dossiers and reviewing about 180 EU Draft Assessment Reports, we have gathered a broad expertise on possible refinement steps in the environmental risk assessment.

For a complete handling of PPP authorisation, we co-operate with our partners.

Our services include:

- Data gap analysis and recommendations for additional testing
- Performing, placing and monitoring of required studies according to GLP
- Preparation of dossiers for the active substance and formulations
- Preparation of the fate (Section 5) and ecotox (Section 6) dossier parts
- Environmental risk assessment and risk refinement
- Project coordination and communication with stakeholders

With regard to biocides ECT's expertise includes exposure and risk assessments by using appropriate Emission Scenario Documents (ESD), the Technical Guidance Document (TGD), the Technical Notes for Guidance (TNsG) and tools such as the European Union System for the Evaluation of Substances (EUSES).

ECT supports its clients during the complete registration process by providing the following services:

- Identification of the product type and specific information requirements
- Data mining, data gap analysis and literature search
- Data management, evaluation and reporting with the IUCLID 5 database software
- Placing, performing and monitoring of required studies according to GLP
- Conducting the environmental risk assessment and specifying exposure scenarios
- Dossier preparation: Doc III (A, B), II (A, B, C) and I
- Other dossier sections can also be handled at ECT or are covered by our partners
- Assessment of substances of very high concern (SVHC): CMR, PBT, vPvB, endocrine disruptors, equivalent level of concern
- Classification, Packaging & Labelling (CLP) according to the Globally Harmonised System (GHS) and EU requirements
- Communication with competent authorities (pre- and post submission)
- Project coordination and communication with all participants
- Submission of the dossier documents to the competent authorities

INDUSTRIAL CHEMICALS

Our Industrial Chemicals-REACH team is specialised in the area of ecotoxicology, fate and behaviour as well as physico-chemical properties of industrial chemicals. Additional areas are covered through the established and successful co-operation with ECT's partners.

Since 2003, ECT has worked on more than 30 SIDS Dossiers (Screening Information Data Set) for high production volume (HPV) chemicals. In cooperation with companies, consortia and competent authorities, SIDS Dossiers, SIARs (SIDS Initial Assessment Reports) and SIAPs (SIDS Initial Assessment Profiles) of more than 10 ICCA/HPV chemicals (incl. chemical categories) have been successfully submitted. Based on our experience in research we are familiar with specific data requirements e.g. related to substances of very high concern (SVHC), persistent, bioaccumulative and toxic (PBT) substances, and endocrine disruptors.

ECT supports its clients during the entire REACH process by offering the following services:

- Identification of information requirements and data gaps
- Data mining and literature search
- Data management, evaluation and reporting with IUCLID 5
- Placing, performing and monitoring of required studies according to GLP
- Conducting the environmental risk assessment and specifying exposure scenarios
- Dossier preparation (e.g. technical dossier, chemical safety report)
- Assessment of substances of very high concern (SVHC): CMR, PBT, vPvB, endocrine disruptors, equivalent level of concern
- Classification, Packaging & Labelling (CLP) according to the Globally Harmonised System (GHS)
- Compiling of Material Safety Data Sheet according to REACH specifications
- Communication with competent authorities (e.g. ECHA)

ECT experts compile dossiers for the environmental risk assessment of human as well as veterinary medicines. Based on our experience in national and international research projects, we can provide scientific expert advice particularly in complex cases of higher tier risk assessment and risk management options.

ECT supports its clients during the pharmaceutical registration procedure by providing the following services:

- Identification of information requirements and data gaps
- Data mining and literature search
- Placing, performing and monitoring of required fate and effect studies according to GLP
- Conducting Phase I and II of the environmental risk assessment for human as well as veterinary pharmaceuticals
- Dossier preparation (CTD Module 1.6)
- Providing expert opinion reports on specific questions regarding effects, exposure and risk management
- Communication with the competent authorities

MIXTURES

The consideration of mixtures of chemical substances in the regulatory assessment of environmental risks is getting into the focus of competent authorities. Recent developments, particularly in the risk assessment of plant protection products and biocides, require the evaluation of mixture toxicity.

Based on the experience obtained in various research projects, we can conduct experimental studies with mixtures as well as apply established mixture toxicity concepts to theoretically predict the effects of mixtures based on the effects of the individual components.

WASTE

The ecotoxicological characterisation of waste is part of their assessment as hazardous or non-hazardous according to the European Waste List (EWL, 2000/532/EC and updates). Based on the experience obtained in organising an international ring test, ECT is perfectly prepared to conduct all tests with waste eluates and solid wastes that are required to evaluate the criterion H14 "ecotoxic". All tests are conducted in compliance with the respective guidelines including the requirements of ISO 17025 (quality assurance).

WORKSHOPS & MEETINGS

Resulting from coordination and participation in large international research projects ECT offer the following services:

- Organisation of workshops and conferences
- Identification and invitation of speakers
- Invitation of participants
- Moderation of the workshop or conference
- Proceedings and summary records

TRAINING ACTIVITIES

TRAINING OF INDIVIDUALS

In co-operation with national and international universities, ECT offers training opportunities for young scientists, engineers and technicians from Germany and abroad. In particular, practical courses and internships, usually ranging from 1 – 6 months, as well as scientific experimental work for Bachelor, Master and Ph.D. degrees are possible.

TRAINING OF GROUPS

Together with about 10 other German institutions (mostly universities), ECT is a partner of the PGS (post gradual studies) initiative in ecotoxicology, jointly organized by the SETAC German Language Branch (SETAC GLB) and the Society of German Chemists (GDCh). In close cooperation with the Rhein-Main University (Wiesbaden-Rüsselsheim) ECT performs courses in terrestrial ecotoxicology. Furthermore, ECT has accumulated experience by participating in international training courses in Portugal, Brazil, Tunisia and Morocco. Participants from universities, environmental agencies and industry learn in these courses about the fate of chemicals in soil, effects of pesticides on non-target arthropods and the principals of quality assurance requirements when performing ecotoxicological tests in the laboratory or the field.

STANDARDISATION

Studies on the effects and the fate of chemicals in the context of legally required assessment procedures are conducted according to international guidelines. ECT actively contributes to the development, standardisation and validation of such methods and guidelines. These activities include the organisation of international ring tests as well as participation in working groups of international standardisation organisations.

ECT contributed significantly to the standardisation of the following methods and guidelines:

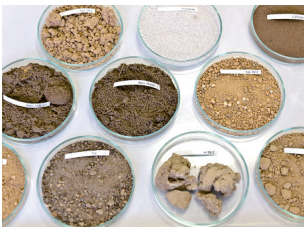
OECD (2008) Detailed review paper on fish-cycle test reproduction test in soil
OECD 315 (2008) Bioaccumulation in sediment-dwelling benthic oligochaetes
OECD 225 (2007) Sediment-water *Lumbriculus* toxicity test using spiked sediment
ISO/DIS 23611-6 (2011) Soil quality - Sampling of soil invertebrates - Part 6: Guidance for the design of sampling programmes with soil invertebrates
OECD 317 (2010) Bioaccumulation in terrestrial oligochaetes
OECD GD 122 (2010) Guidance document on the determination of the toxicity of a test chemical to the dung beetle *Aphodius constans*
OECD 226 (2008) Predatory mite *(Hypoaspis (Geolaelaps) aculeifer)* reproduction test in soil
OECD 228 (2008) Dung flies *(Scathophaga stercoraria, Musca autumnalis)* laboratory tests
ISO 17512 (2007) Soil Quality - Avoidance test with earthworms *(Eisenia fetida/andrei)*
ISO 23611-3 (2007) Soil quality - Sampling of soil invertebrates - Part 3: Sampling and soil extraction of enchytraeids
ISO 23611-1 (2006) Soil quality - Sampling of soil invertebrates - Part 1: Hand-sorting and formalin extraction of earthworms
OECD GD 54 (2006) Breakdown of organic matter in litter bags in the field
OECD 222 (2004) Earthworm Reproduction Test *(Eisenia fetida/Eisenia andrei)*
OECD 220 (2004) Enchytraeidae reproduction test
ISO 16387 (2004) Soil Quality – Effects of pollutants on Enchytraeidae *(Enchytraeus sp.)*
ASTM 1676 (2004) Standard guide for conducting laboratory soil toxicity or bioaccumulation tests with the lumbricid earthworm *Eisenia fetida* and the enchytraeid potworm *Enchytraeus albidus*
ISO 22030 (2004) Chronic toxicity in higher plants

ISO (INTERNATIONAL ORGANISATION FOR STANDARDISATION)

ISO and its national partners (e.g. the German Institute for Standardisation DIN) are non-profit associations which standardise, validate and publish standards for technical equipment and experimental methods. ECT's member of staff are active in the Committees ISO/TC 190/SC 4 „Biological methods", especially in the Working Groups "Effects on soil fauna" (WG 2), "Effects on soil flora" (WG 3) and "Effects on soil microorganisms (WG 4) and ISO/TC 190/SC 7 (Working Group 3 "Ecotoxicological characterisation of soils and soil materials" and Working Group 8 "Bioavailability").

OECD (ORGANISATION FOR ECONOMIC CO-OPERATION AND DEVELOPMENT)

An important aspect of OECD's Chemicals Safety Program is to coordinate the development, standardisation, validation and publication of Chemicals Testing Guidelines. In this process expert groups can be established and members of these expert groups are nominated by the National Coordinators of each member country. Staff members of ECT have been nominated as experts for the standardisation and validation of various guidelines in the test guideline section "Effects on Biotic Systems". (e.g. testing of endocrine disrupting chemicals with fish and invertebrates).



Our research activities range from rather basic to applied aspects in different areas of ecotoxicology and ecology. We investigate cause-effect relationships of chemicals at various levels of biological organisation, ranging from molecular structures, physiological functions, individuals, and populations to (model) ecosystems. In more applied research projects, we improve existing or develop new testing methods to assess potential impacts of chemicals in the environment. Another field of our research relates to the improvement of existing environmental risk assessment schemes and the provision of scientific background knowledge to improve regulatory decisions. In addition, we participate in ecological studies in the context of nature protection and soil biological monitoring activities, in Europe and in North and South America (especially Brazil).

We frequently operate as partner or coordinator of national and international consortia. Our partners in research are universities, national and international research institutions, governmental organisations and companies from the chemical industry.

We actively participate in the Society of Environmental Toxicology (SETAC) and in the "Fachgruppe Umweltchemie und Ökotoxikologie der Gesellschaft für Deutsche Chemiker (GDCh)" [Working Group Environmental Chemistry and Ecotoxicology of the German Chemical Society (GDCh)].

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