

Annex I: Project Progress Summary

Project Progress Summary

<p><u>Section 1: PROJECT IDENTIFICATION</u> Information to be provided for project identification</p>	<p>NOT CONFIDENTIAL</p>	
<p>Title of the project</p> <p>ENVIRONMENTAL AGENTS SUSCEPTIBILITY ASSESSMENT UTILISING EXISTING AND NOVEL BIOMARKERS AS RAPID NON-INVASIVE TESTING METHODS</p>		
<p>Acronym of the project EASYRING</p>		
<p>Type of contract RTD</p>		
<p>Total project cost (in euro)</p> <p>2351,056 €</p>		
<p>Contract number</p> <p>QLK4-2002-02286</p>	<p>Duration (in months)</p> <p>36 Months</p>	<p>EU contribution (in euro)</p> <p>1890,209 €</p>
<p>Commencement date</p> <p>01/01/2003</p>		<p>Period covered by the progress report</p> <p>1 January 2003 – 31 December 2004</p>
<p><u>PROJECT COORDINATOR</u></p>		
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<p>Key words (5 maximum - Please include specific keywords that best describe the project.).</p> <p>Endocrine disrupters, Biomarkers, Risk assessment, <i>in vitro</i>, dipstick</p>		
<p>World wide web address (the project's www address)</p> <p>WWW.easyring.org</p>		

List of participants Provide all partners' details including their legal status in the contract i.e., contractor, assistant contractor (to which contractor?).

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Annex I: Project Progress Summary

Section 2: Project Progress Report

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(2 pages maximum.. Use short sentences. Be factual. Avoid technical terms as much as possible)

Objectives:

The major objectives and achievements expected for EASYRING are:

- Rapid, non-invasive test development for the identification of specific biomarkers for endocrine disrupting chemicals in the mucus of aquatic species.
- Novel functional biomarker(s) for the identification of endocrine disrupting chemicals.
- Production and validation of new enzyme linked immunosorbent assays (ELISAs) for the detection of new biomarkers in plasma, mucus and tissues of aquatic species.
- Improvement of *in vitro* test for the screening of endocrine disrupting chemicals.
- Development of analytical methods in GC-MS and LC-MS for selected endocrine disrupting chemicals in environmental water and biota.
- Short/long term and low level exposure effects of selected chemicals and mixture in low vertebrates.
- Short/long term and low level exposure effects of selected chemicals and mixture, through food and water chain, in small mammals.
- QSARs development for the prediction of chemicals able to elicit endocrine disruption.
- QAARs development to extrapolate the responses of the different experimental models.

Results and Milestones:

- 1) Establishment of retinol binding protein and transthyretin as new biomarker in *Xenopus* (M1)
- 2) Identification of established biomarker proteins (Vtg/) in mucus of exposed fish (M8)
- 3) Partial cloning of DMRT1-like gene of carp and related real time-PCR analytic protocol (M13).
- 4) Development of assays on pre-implantation embryos (M21).
- 5) Collection of data for table of anomalies in early development stages of studied species, caused by long-term exposures to selected chemicals and their mixtures
- 6) Collection of data for table of morphological alterations caused by selected chemicals and mixtures after short and long-term exposures
- 7) Collection of data for table of biochemical alterations caused by selected chemicals and mixtures after short and long-term exposures Table of biochemical alterations caused by selected chemicals and mixtures after short and long-term exposures
- 8) The development of models (QSARs) for the prediction of the endocrine disruption capability of compounds. Comparison of different methodologies for the prediction of activity (M27)
- 9) Website

Benefits and Beneficiaries:

- Improvement of the standard validated tests for risk assessment (*beneficiaries*: policy makers, scientist, national and regional environmental agencies, environmental quality monitoring agencies and laboratories)
- Development of new non invasive test methods (*beneficiaries*: policy makers, scientist, national and regional environmental agencies, environmental quality monitoring agencies and laboratories)
- Improvement of *in vitro* test systems with alternative screening and testing protocols (*beneficiaries*: scientist, environmental quality monitoring agencies and laboratories)
- Improvement of chemical analytical method and their application (beneficiaries: scientist, environmental quality monitoring agencies and laboratories)
- Improvement of knowledge on Cytotoxic and teratogenic effects of endocrine disrupters/modulators and quantification of exposures in mammals (cause-effects relationships investigation) (*beneficiaries*: policy makers, national and regional environmental and health agencies, scientist).

Future Actions (if applicable):

For the future, activities will be performed following the original timetable foreseen in the technical annex. Even though, due to the problems arisen during the first year of work especially related to biomarker genesis, the Consortium set up strategies in order to solve the problem and close the gap.

As requested by the referees the Consortium has been set up a dissemination plan. The choice will be to prefer interdisciplinary article produced among partners, in order to obtain a wider view point. So these "macro argument" should be more comprehensive at different scientific approaches. Scientific groups will be engaged to increase the dissemination of the emergent data within the end of 2005.

The third year activities in field will include specific studies on the target species on the two sampling points, located upstream and downstream the Lambro River confluence. In particular, the activities carrying on are aimed to: study the diet composition of the target species through the analysis of the stomach contents, determine the linear growth rate through the scalimetric method that allows the age determination and determine the absolute and relative fecundity through the count of eggs present in ovary sub-samples. The prototype of dipstick for the detection of Vtg in the mucus of fish will be checked on-site in the field studies planned during these surveys in Spring 2005.

Regarding modeling starting by month 30 the database will be formally closed. At this time will be contain quantitative and qualitative data from various endpoints. In addition it will include a selection of physico-chemical properties and descriptors that have been found to be important in the modelling process. The database will be placed on the Project internet page. The possibility of converting the database using an internet mark-up language (XML) in collaboration with the UK EPSRC project "Pythia" (University of Bradford) will be explored.